UNIVERSIDADE FEDERAL DO ESTADO DO RIO DE JANEIRO - UNIRIO
CENTRO DE CIÊNCIAS BIOLÓGICAS E DA SAÚDE - CCBS
INSTITUTO DE BIOCIÊNCIAS - IBIO
PROGRAMA DE PÓS-GRADUAÇÃO EM CIÊNCIAS BIOLÓGICAS: BIODIVERSIDADE NEOTROPICAL - PPGBIO

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Aspectos ecológicos da distribuição dos nematódeos em praias arenosas do Rio de Janeiro

Rio de Janeiro
2019

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Dissertação apresentada ao Programa de Pós-Graduação em Ciências Biológicas (Biodiversidade Neotropical) da Universidade Federal do Estado do Rio de Janeiro como requisito parcial para obtenção do título de Mestre.

Orientadora: Prof. Dra. Tatiana Fabricio Maria

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Aprovada em 11 de fevereiro de 2019.

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Harley Costa Santos, Gabriel
H237 Aspectos ecológicos da distribuição dos nematódeos em praias arenosas do Rio de Janeiro / Gabriel Harley Costa Santos. -- Rio de Janeiro, 2019.
63 f.

Orientador: Tatiana Fabricio Maria.
Dissertação (Mestrado) - Universidade Federal do Estado do Rio de Janeiro, Programa de Pós-Graduação em Ciências Biológicas, 2019.

1. Ecologia Bêntica. 2. Praias Arenosas. 3. Biomonitoramento. 4. Nematofauna. 5. Granulometria. I. Fabricio Maria, Tatiana, orient. II. Título.

À minha avó, Agostinha, que merecia ter sido a cientista que quis ser.

AGRADECIMENTOS

Agradeço à minha mãe, Anilda Gomes da Costa, que com muito amor, dedicação, ética e garra, me ajudou e inspirou a ir atrá de tudo que já conquistei. Obrigado por ter me dado a liberdade para eu ser quem realmente sou e me encorajado a ir atrás dos meus sonhos, mesmo sem entender exatamente quais eles são. Eu sou o filho mais orgulhoso do mundo.

Agradeço também à minha irmã, Gabriela Gomes, por toda ternura e por todo cuidado durante todas as fases da minha vida. Sempre aprendi muito te tendo como exemplo de irmã e fico muito feliz em continuarmos nossa parceria de vida mesmo quando o acaso nos distanciou tanto.

Agradeço à Professora Tatiana Fabricio Maria, minha orientadora, por ser o exemplo de profissional que eu quero me tornar e por toda disponibilidade oferecida nesses anos de parceria. Sempre tive a sorte e a honra de estar cercado por mulheres incríveis e talentosas, mas foi trabalhando com você que foi renovada a minha esperança em várias coisas em que eu já estava desacreditado. Você me inspira de formas que em muito transcendem as barreiras do mundo acadêmico. Obrigado pela confiança e pela oportunidade.

Agradeço ao Laboratório de Ecologia Bêntica (LEB) e todos que compuseram nossa equipe durante os últimos anos: Alyne Cristine Corrêa Martins, Bruna Miranda Gomes Lesaige, Camila Silva do Rosário, Leo Afonso Perez, Letícia Maria Monteiro Soares, Luís Otávio Vítor de Lima e Mayara Faustino de Sá. Este projeto não poderia ter sido realizado sem o esforço de todos nós desde as etapas de coleta até a identificação dos nematódeos. Agradeço em especial à Raíssa Vieira Côrrea, pelo suporte, ajuda e parceria desde o meu começo no LEB.

Agradeço à Universidade Federal do Estado do Rio de Janeiro (UNIRIO), por ter me acolhido nos últimos sete anos, pelas oportunidades oferecidas e pelas bolsas de iniciação científica fornecidas. Agradeço ao Programa de Pós-Graduação em Ciências

Biológicas: Biodiversidade Neotropical (PPGBIO) por me permitir integrar seu corpo discente e confiar na execução deste trabalho. À Fundação Carlos Chagas Filho de Amparo à Pesquisa do Estado do Rio de Janeiro (FAPERJ), pelo suporte financeiro para a realização do projeto “Impactos antrópicos em praias arenosas fluminenses: uma abordagem ecológica utilizando bioindicadores naturais”, do qual as amostras para esse estudo foram obtidas. Agradeço também ao Prof. Dr. Ricardo Silva Cardoso, pelo convite para nosso grupo de pesquisa participar do projeto.

À Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES) por me conceder a bolsa de estudos durante a pós graduação e pela oportunidade de participação no Programa Jovens Talentos Para a Ciência durante a graduação, incentivos mais do que necessários para que os frutos desse trabalho fossem possíveis. Agradeço também ao Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) pela oportunidade de participação no Programa Ciências Sem Fronteiras, onde pude viver algumas das experiências mais importantes e enriquecedoras da minha vida acadêmica até agora.

Agradeço ao Msc. Michel Almeida, do Laboratório de Ecologia Marinha da UNIRIO e ao Prof. Dr. Ricardo Pollery, do Laboratório de Biogeoquímica da UFRJ, por fornecer a estrutura e auxiliar nas análises granulométricas realizadas. À Dra. Tatiana Cabrini, pela disponibilidade e por fornecer informações que muito incrementaram na caracterização das praias deste estudo. À Prof. Dra. Natascha Krepsky, do Laboratório de Microbiologia das Águas da UNIRIO, por fornecer os dados referentes aos coliformes e à Iracema Prestes Brandão, que auxiliou nas coletas e nas análises dessas amostras. Ao meu colega Guilherme Maricato, por gentilmente me auxiliar na elaboração dos mapas utilizados e à Msc. Ana Clara Franco, pela ajuda na metodologia utilizada para estimar a densidade humana. Agradeço também à minha amiga Haelly Dragnev pelos diversos suportes gráficos prestados ao longo desse projeto.

Por fim, agradeço aos amigos e colegas que dividiram comigo os mais profundos e diversos aprendizados e experiências durante os últimos anos, em especial ao Diogo, que me ajudou a desbravar (e me manter) em terras cariocas e fez com que tudo fosse sempre mais leve.

RESUMO

Os nematódeos são os organismos mais representativos nos sedimentos marinhos, chegando a representar 90% da comunidade meiofaunística em ambientes costeiros. Esses organismos podem ser encontrados em um enorme espectro de condições ambientais, desde ambientes pristinos a altamente impactados, dependendo apenas de uma fonte de alimento para se estabelecer. Nas praias arenosas, os nematódeos são o principal grupo encontrado na meiofauna intersticial, sendo as características granulométricas tidas como as principais variáveis ambientais responsáveis pela distribuição desses organismos nesses ambientes. Além disso, as distintas características fisiológicas e morfológicas dos nematódeos os tornam capazes de produzir diferentes respostas ecológicas a níveis de organismos, populações e comunidade, o que vem sendo visado como um potencial uso desse grupo como bioindicadores. O biomonitoramento com o uso desses organismos deve sempre estar relacionado às características físico-químicas do ambiente e os impactos analisados devem ser bem delineados para que os índices que utilizam esses grupos como indicadores sejam interpretados de forma correta. No Rio de Janeiro, a caracterização da nematofauna e sua relação com os aspectos ecológicos encontrados nas praias fluminenses seguem inexplorados, apesar da grande importância desses ambientes para a recreação e o turismo no Estado. Dessa forma, o capítulo único presente nesta dissertação, submetido para a revista Estuarine, Coastal and Shelf Science, trata da avaliação das respostas ecológicas dos nematódeos encontrados em praias arenosas do Rio de Janeiro sob diferentes níveis de impactos antrópicos, cujo objetivo foi identificar as principais variáveis responsáveis pela formação das comunidades nesses ambientes e aplicar os índices que utilizam esses organismos como bioindicadores. Sete praias arenosas do Estado (Barra da Tijuca, Botafogo, Copacabana, Fora - Paraty, Fora - Urca, Guaxindiba e Restinga da Marambaia) foram selecionadas de acordo com os diferentes tipos de impactos a que estão sujeitas e amostradas durante o verão, na alta temporada turística. Amostras para extração e identificação dos nematódeos e caracterização ambiental (granulometria e matéria orgânica) foram obtidas em 30 pontos de coleta dispostos em três transectos fixos e equidistanes em cada uma das praias, na zona entremarés. Também foi realizada a aferição das principais variáveis físico-químicas em cada uma delas através de uma sonda multiparamétrica e a caracterização microbiológica para a quantificação dos coliformes termotolerantes. A granulometria foi analisada através de um analisador de partículas a laser e o teor de matéria orgânica estimado através da pesagem e queima do sedimento. Os nematódeos foram contados e identificados através de chaves pictoriais, cujos resultados foram posteriormente utilizados para o cálculo dos índices biológicos. Os resultados apontaram para a presença de 100 diferentes gêneros de nematódeos encontrados em todas as praias, com diferenças significativas na riqueza e densidade entre as praias. Os índices biológicos aplicados separadamente não apresentaram respostas claras quanto ao *status* ambiental das praias arenosas. O grau de seleção do sedimento foi a variável que melhor explicou a distribuição dos nematódeos das praias analisadas em conjunto, apresentando inclusive uma importância superior ao tamanho médio do grão, que, geralmente, representa a variável mais estudada e que explica a distribuição dos nematódeos. Observou-se também a preferências de alguns gêneros identificados a determinados espectros de grau de seleção, o que levou à conclusão de que as características granulométricas representam maior importância na distribuição dos nematódeos das praias arenosas do Rio de Janeiro quando comparados aos impactos antrópicos a que essas estão submetidas. Além disso, este manuscrito representa uma importante base de dados para futuros trabalhos de monitoramento com o uso dos nematódeos por representar a primeira caracterização de pelo menos seis praias do litoral fluminense.

Palavras-Chaves: Biomonitoramento, Praias arenosas, Ecologia Bêntica, Nematódeos.

ABSTRACT

Nematodes are the most representative organisms in marine sediments, accounting for 90% of the meiofauna assemblage in coastal environments. Individuals of this group have already been found in a wide range of environmental conditions, from pristine to highly impacted environments, only relaying on a food source to establish. In sandy beaches, nematodes are the main group found in interstitial meiofauna, with the granulometric characteristics being the main responsible for their distribution in these environments. Besides that, nematodes wide morphological and physiological characteristics make them capable of producing different ecological responses at individual, population and community levels that has been viewed as a potential use of this group as bioindicators. Using nematodes for biomonitoring requires that physicochemical characteristics of the environment are widely measured and impacts need to be well delineated, so the biological indexes that use this group as indicators are rightfully interpreted. Despite the great importance of these environments for recreation and tourism in Rio de Janeiro, the nematofauna characterization and its relationship to the ecological aspects found in the beaches of the State remain unexplored. Thus, the unique chapter presented in this dissertation, submitted to the journal Estuarine, Coastal and Shelf Science, deals with the evaluation of the ecological responses of nematodes found on sandy beaches of Rio de Janeiro under different levels of anthropic impacts. The aim was to identify the main variables responsible for the formation of nematode assemblages in these environments and to apply the biological indexes that use this group as bioindicators. Seven sandy beaches from Rio de Janeiro state (Barra da Tijuca, Botafogo, Copacabana, Fora - Paraty, Fora - Urca, Guaxindiba e Restinga da Marambaia) were selected according to the different types of impact they are facing and sampled during the summer, on the high touristic season. Samples for nematode extraction and environmental characterization (granulometry and organic matter content) were obtained from 30 sampling stations on each one of the beaches, placed on three equidistand transects located at the intertidal zone. The main environmental physicochemical variables that were also measured at each beach with a multiparametric probe and the microbiological characteristics measured for thermotolerant colifotms quantification. Granulometry was analyzed through a laser particle size analyzer and organic content estimated through the weighting and burning of sampled sediment.

Nematodes were counted and identified using pictorial keys, with results being later used for the calculation of the biological indexes. Results showed the presence of 100 different nematode genera on all beaches combined, with significantly differences on richness and density between them. Biological indexes individually did not present clear answers about the environmental *status* of the sandy beaches. The sorting coefficient was the variable that best explained nematode distribution on the beaches analyzed altogether, presenting higher importance when compared to the mean grain size, which is the most commonly studied variable and with the most known associations. It was also shown that some nematode genera have preferences to certain sorting coefficient spectrums, what led to the conclusion that environmental characteristics represents higher importance on nematode distribution on sandy beaches from Rio de Janeiro when compared to the anthropic pressures that these environments face. This work also represents an important database for future nematode monitoring works as it represents the first assemblage characterization of at least six beaches along the coast of Rio de Janeiro.

Keywords: Biomonitoring, Sandy beaches, Benthic ecology, Nematodes.

LISTA DE ILUSTRAÇÕES

LISTA DE TABELAS

SUMÁRIO

1. INTRODUÇÃO

1.1 Praias arenosas, meiofauna e biomonitoramento

Representando cerca de dois terços dos ambientes costeiros do mundo, as praias arenosas estão entre os ecossistemas mais amplamente distribuídos e podem ser localizadas na maioria dos países tropicais e temperados (MCLACHLAN & BROWN, 2006; SCHLACHER, 2015). Esses ecossistemas representam um importante elo da sociedade moderna com os oceanos, já que suportam a realização de diversas atividades recreativas e econômicas e são, inclusive, o principal atrativo turístico de diversas cidades costeiras (HOUSTON, 2008).

Embora sejam ecossistemas dinâmicos e oferecerem um variado espectro de habitats, influenciados por fatores físicos como as ondas, as correntes marinhas, o vento e o regime de marés (BROWN & MCLACHLAN, 2002), as praias arenosas foram por muito tempo tidas como desertos biológicos e pouco ainda é conhecido a respeito das relações ecológicas ocorridas nesses locais, estando os principais estudos em áreas costeiras concentrados nos costões rochosos e com abordagens sobre a ictiofauna desses ambientes (MCLACHLAN & BROWN, 2006).

As praias arenosas são o abrigo de diversos representantes da megafauna, que engloba as aves marinhas e grandes crustáceos, e da macrofauna, representada pelos tatuís, anfípodes e outros pequenos invertebrados. Outro grupo de organismos também presente nas praias arenosas é a meiofauna, que mesmo invisíveis a olho nu, estão altamente adaptados para viver entre os grãos de sedimento. A meiofauna não representa um grupo taxonômico, incluindo organismos de mais de 25 táxons com características fisiológicas e morfológicas distintas, mas que permitem que os indivíduos habitem a película d’água formada entre os grãos de areia da praia (GIERE, 2009).

A classificação da meiofauna se dá de acordo com a metodologia de extração dos organismos que a compõem, compreendendo os que passam por uma malha de 500µm (limite superior) e são retidos por uma malha de 31 µm (limite inferior) (GIERE, 2009). O grupo representa um importante elo no fluxo de energia dos sistemas bentônicos, atuando como consumidor primário dos organismos microbentônicos e como fonte de alimento para jovens peixes e crustáceos (COULL, 1990; ARGEIRO, 2009).

Dos diversos grupos componentes da meiofauna, como os copépodes, turbelários, oligoquetas, tardígrados e poliquetas, os nematódeos se destacam como o grupo mais abundante, chegando a representar entre 60 a 90% da comunidade meiofaunística em sedimentos marinhos

(COULL, 1999). Esses organismos podem ser encontrados em todos os tipos de sedimento, sob quaisquer condições climáticas e em habitats cujo status ambiental variam de intocados a altamente poluídos, dependendo apenas da presença de uma fonte de carbono orgânico disponível (BONGERS & FERRIS, 1999).

Em praias arenosas, a distribuição da meiofauna e, consequentemente, dos nematódeos, é influenciada principalmente por variáveis ambientais como a granulometria, precipitação, quantidade de matéria orgânica no sedimento, oxigênio disponível e a temperatura (GIERE, 2009; STEYAERT et al., 2007). As características granulométricas são destacadas como as mais importantes, onde o tamanho do grão e o grau de seleção são os mais estudados e associados à dominância de certos gêneros e famílias (FONSECA et al. 2014; URBAN-MALINGA et al., 2014). O tamanho médio dos grãos e o grau de seleção influenciam no nível de heterogeneidade do sedimento, que está diretamente relacionado à quantidade de nichos ecológicos disponíveis para as espécies bênticas (ABSALÃO et al., 2006). A interpretação dos efeitos dessas variáveis pode ser complementada com outros fatores granulométricos como a assimetria, que indica quais frações granulométricas mais contribuem para a diversidade do sedimento e a kurtosis, que revela informações a respeito do transporte e da sedimentação das diferentes frações granulométricas (FOLK & WARD, 1957, ABSALÃO et al., 2006).

A densidade de indivíduos e a riqueza de espécies já mostraram estar associadas ao tamanho do grão de sedimento (VANAVERBEKE et al., 2011), porém existem poucas evidências de que essa variável por si só seja a principal responsável pela distribuição dos organismos intersticiais (SNELGROVE & BUTMAN, 1994). O regime hidrodinâmico das praias arenosas faz com que o tamanho do grão covarie com outros fatores chave como o teor de matéria orgânica, a porosidade e a abundância/composição microbial do sedimento, o que é exemplificado pelo fato de muitas espécies não estarem associadas a um único tipo de sedimento (SNELGROVE & BUTMAN, 1994).

Em conjunto com as características do ambiente, o grupo Nematoda também responde de diferentes formas a diferentes tipos de impactos antrópicos, sejam estes de natureza química ou física (MOORE & BETT, 1989). Essa capacidade de resposta aliada às características gerais dos representantes da meiofauna (ciclo de vida curto, alta abundância de organismos nos sedimentos marinhos, tamanho diminuto e organização corporal simples) demonstram a possibilidade de utilizar os nematódeos como bioindicadores de impactos humanos (MORENO et al., 2008).

O uso dos nematódeos como indicadores de qualidade ambiental vem sendo bastante visado por apresentar vantagens em relação a grupos tradicionalmente utilizados, como a macrofauna (KENNEDY & JACOBY, 1999). A amostragem da meiofauna é mais simples e, com pequenos volumes de sedimento, é possível coletar um número suficiente de organismos para realizar análises estatísticas robustas e pertinentes (KENNEDY & JACOBY, 1999). A desvantagem no uso da meiofauna e dos nematódeos como bioindicadores está principalmente na dificuldade de identificação dos organismos, principalmente pelo seu tamanho diminuto, e na interpretação dos dados, já que, além dos impactos a que estão sujeitos, diversas variáveis ambientais estão envolvidas na distribuição desses organismos (FONSECA et al., 2014; WARD, 1975).

Alguns estudos já realizaram com sucesso o uso dos nematódeos como indicadores de diversos tipos de impactos antrópicos, através da utilização e da criação de diversos índices cujo objetivo é de caracterizar o *status* ecológico de um ambiente com base na nematofauna presente (GHESKIERE et al., 2005; MORENO et al., 2011; MORENO et al., 2009) . Podem ser destacados: (1) densidade de organismos; (2) índices de diversidade, tais como os índices de Margalef, diversidade de Shannon, riqueza de espécies, diversidade esperada e diversidade beta; (3) índice de maturidade (BONGERS & HAAR, 1990) e (4) índice de diversidade trófica (HEIP et al., 1985). Além disso, a própria presença/ausência de determinados gêneros é tida como um indicativo das condições deste ambiente (MORENO et al., 2011).

Apesar da existência desses índices e dos avanços no uso dos nematódeos como bioindicadores, a maior parte dos estudos relacionados à nematofauna nas praias arenosas do Brasil possuem caráter descritivo, que objetiva fazer o levantamento dos gêneros de nematódeos encontrados em determinadas praias (SILVA et al., 1991; ESTEVES et al., 1995; ESTEVES et al., 1997; WANDENESS et al., 1997; ESTEVES & SILVA, 1998; ESTEVES et al., 1998; SILVA et al., 1999; OLIVEIRA & SOARES-GOMES, 2003; ESTEVES, 2004; ALBUQUERQUE et al., 2007; MARIA et al., 2008; MARIA et al., 2013). Não obstante, a influência das características físico-químicas na distribuição da nematofauna possui diversas lacunas que podem oferecer importantes informações a respeito da autoecologia dos diversos gêneros desse grupo. Ao realizar o estudo desse grupo nesses ambientes de forma ecológica, com as devidas associações entre a comunidade dos nematódeos (através da aplicação de índices biológicos) e os fatores físico-químicos e microbiológicos, o *status* ambiental das praias arenosas pode se tornar mais fácil de ser visualizado e ajudar a destacar a necessidade de conservação das praias arenosas, que categorizam ambientes impactados por diversas ações humanas, como, por exemplo, o despejo de esgoto, acúmulo de lixo e pisoteio (CARDOSO et al., 2016).

1.2 Escolha do Tema

No Rio de Janeiro, apenas seis praias tiveram a comunidade meiofaunística e/ou o levantamento dos gêneros de nematódeos publicados, sendo elas a Praia da Bica, Praia de Coqueiros, Praia do Bananal, Praia de Charitas, Praia Vermelha e Restinga da Marambaia, que foram analisadas em 12 diferentes estudos (MARIA et al., 2016). Nesse estudo, sete praias tiveram sua nematofauna avaliada, estando tais praias submetidas a diferentes graus de impactos antrópicos, o que sugere que elas possuam diferentes *status* ecológicos.

Nas praias de Copacabana e Barra da Tijuca, ambas localizadas no município do Rio de Janeiro, os impactos advindos do uso humano ocorrem quase que durante o ano todo, devido à alta frequência de turistas e moradores (CARDOSO et al., 2016). Ainda assim, a nematofauna dessas praias de alta importância socioeconômica encontra-se desconhecida ou pouco estudada. O mesmo ocorre nas praias de Fora em Paraty e Guaxindiba em São Francisco do Itabapoana, que são utilizadas principalmente durante o verão, na alta temporada turística, mas com baixa frequência de banhistas durante o restante do ano. Em contraste, as praias da Restinga da Marambaia e de Fora - Urca, apesar de também estarem localizadas na cidade do Rio de Janeiro, são consideradas pouco impactadas e com elevado status de preservação, já que possuem acesso restrito por estarem localizadas em áreas militares (CARDOSO et al. 2016). Na praia de Botafogo, que também é pouco frequentada por banhistas, a elevada contaminação causada pelo despejo de dejetos orgânicos (FISTAROL et al., 2015) é o que torna esse ambiente diferente dos anteriores, estando essa praia em um nível muito superior de degradação.

A comparação da comunidade nematofaunística desses ambientes que possuem impactos tão distintos entre si e suas relações com os fatores medidos em cada uma das praias pode permitir verificar quais são as variáveis ambientais mais importantes na distribuição da nematofauna nas praias arenosas do Estado e verificar se exitem gêneros com maiores afinidades a determinadas características do ambiente. Além disso, essa análise também pode revelar respostas ecológicas que potencialmente auxiliarão no monitoramento das praias arenosas, através da aplicação dos índices biológicos existentes e da caracterização da nematofauna que segue praticamente inexplorada para a maioria das praias do Rio de Janeiro e do Brasil.

1.3 Objetivo

O objetivo deste projeto foi avaliar a nematofauna de sete praias arenosas do estado do Rio de Janeiro, de forma que fosse possível verificar quais as variáveis ambientais mais importantes na distribuição dos organismos e aplicar os índices que utilizam o grupo Nematoda como bioindicadores para verificar o *status* ambiental desses ambientes.

Os objetivos específicos foram:

- Caracterizar a nematofauna de acordo com a identificação dos gêneros de nematódeos encontrados.
- Aplicar os índices de densidade, riqueza (S), diversidade beta, diversidade trófica e maturidade de forma que fosse possível relacionar os resultados com os diferentes impactos antrópicos presentes em cada praia.
- Buscar relações entre os parâmetros físico-químicos e microbiológicos com a distribuição da nematofauna nesses ambientes.

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Capítulo I

Bioindicators or sediment relationships: Evaluating ecological responses from sandy beach nematodes

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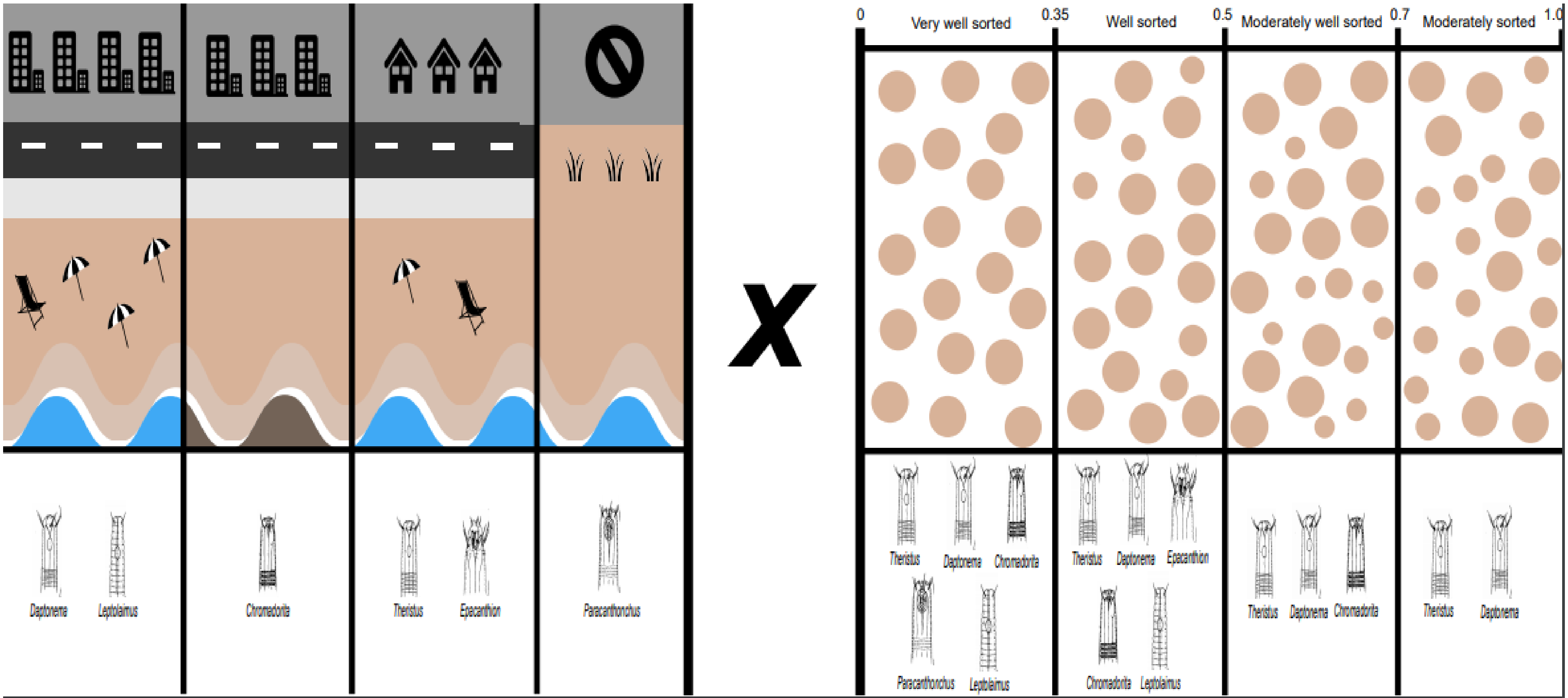
Abstract

Sandy beaches from tropical countries are currently facing many challenges due the rapid urban development and its impacts on coastal areas. Despite their aesthetic, recreational and ecological importance, these environments are neglected when it comes to assessment and conservation measures, which can be done in several ways, including biomonitoring. Nematodes are amongst the most suitable organisms to be used as ecological indicators in sandy beaches due their high abundance on marine sediments and its rapid responses to different sorts of impacts. We analyzed the nematode assemblages and the environmental variables from seven sandy beaches under different types of pressure from Rio de Janeiro state, in Brazil, and applied the biological indexes that use nematodes as bioindicators in order to evaluate differences on the ecological *status* of these beaches. Sampling took place in summer of 2015, during the spring low tide. Physicochemical characteristics, microbiology and granulometry were measured alongside human density on each of the beaches and linked to nematode distribution found on the intertidal zone. Botafogo beach, already known for being highly affected by sewage disposal, showed high values for organic matter content and the least diverse assemblage, with high dominance from genus *Chromadorita*. Despite the higher human density, most urban and touristic beaches (Barra da Tijuca and Copacabana) did not show distinct values for the diversity measures when compared to the most preserved beaches (Restinga da Marambaia and Fora - Urca). While the applied biological indexes did not show clear answers concerning the ecological health of the studied sites; the sediment sorting coefficient was the variable that best explained the nematode distribution and some genera are associated to certain spectrums with this variable. For instance, opportunist genera such as *Daptonema* and *Theristus* were found on broader spectrums of sorting coefficient while predators with bigger body diameters showed a more restricted distribution. As the mean grain size is still considered the main variable on nematode distribution for sandy beaches, this study reveals that the sorting coefficient also gives important answers for the nematode distribution.

Keywords: Biomonitoring; Sorting Coefficient; Free-living Nematodes; Ecological Indicators

Graphical Abstract



1. Introduction

Sandy beaches represent two thirds of all coastal ecosystems of the world, being one of the main touristic and recreational attractions of most coastal cities located in tropical and temperate countries (MCLACHLAN & BROWN, 2006). Their wide use and socio-economic importance displayed on our society contrasts with the lack of assessments and measures to ensure the conservation of these environments when compared to other shoreline areas (DEFEO & MCLACHLAN, 2005). The rapid urban development that has been happening around coastal cities affects the beach ecosystem in several ways (SCHLACHER et al., 2007; MCLACHLAN et al., 2013), which it can become highly degraded in short periods of time if impacts are not measured and/or controlled. In recent years, biomonitoring has become one of the most validated techniques for impact assessment, since the use of living organisms offers integrated responses from biotic and environmental variables present on an ecosystem due to their capacity of adaptation (CASAZZA et al., 2002).

Nematodes are amongst the most suitable organisms used in biomonitoring studies as they represent the most abundant group of metazoans from marine sediments and can be found on all types of environments, independent of its ecological *status* and under any climatic conditions (ZEPILLI et al., 2015; CARRIÇO et al., 2013). Some key features, such as their short life cycle, high abundance, small size and simple body structure (GIERE, 2009) also makes nematodes more suited to be used as bioindicators when compared to traditionally studied organisms from sandy beaches, such as the macro- and megafauna (KENNEDY & JACOBY, 1999; ALVES et al., 2013), especially because of the high densities on small but statistically adequate volumes of samples. These organisms also give different responses to different sorts of impact, both qualitatively and quantitatively, with substitution of assemblages and/or density fluctuations due to a determined local stressor (MOORE & BETT, 1989; MORENO et al., 2011).

Besides anthropic pressures, nematode assemblage distributions are mainly influenced by environmental variables such as precipitation, amount of available organic matter, temperature, dissolved oxygen, salinity and granulometric characteristics such as the mean grain size and the sediment sorting coefficient (BONGERS & FERRIS, 1999, FONSECA et al., 2014; GHESKIERE et al., 2005). Many benthic studies have linked granulometry features, mainly the mean grain size, to the distribution and dominance of some nematode families (e.g. URBAN-MALINGA et al., 2014; MARIA et al., 2012) and it is usually postulated that they are more abundant on finer sediments (GIERE, 2009; GHESKIERE et al., 2004). Diversity and species richness have also found to be higher on more poorly sorted sediments as it provides environmental suitable spaces for nematodes (WARD, 1975).

Studies focusing on characterization and associations among nematode assemblages and environmental factors have been reported for a limited number of Brazillian sandy beaches (MARIA et al., 2016). However, these assessments do not explain the ecological *status* of these environments and many tools created for biomonitoring sandy beaches using nematodes are not applied. Indexes such as the Shannon diversity, beta diversity, the maturity index (BONGERS & HAAR, 1990), the index for trophic diversity (HEIP et al., 1985) and the presence/absence of a few genera itself are some of the techniques that could successfully be used to assess the environmental quality of sandy beaches (MORENO et al., 2011).

The aims of the present study were to: (1) evaluate the ecological *status* of seven different sandy beaches from Rio de Janeiro's coastline and characterize them according to their nematode assemblages, (2) apply some of the existent indexes for ecological assessment and (3) try to find relationships of nematode genera occurrence with environmental variables found on each of these beaches. Our null hypothesis was that differences on anthropic impacts among the sampled beaches would best explain nematode distribution on them while the alternative hypothesis was that nematode assemblage distribution was mainly affected by other variables other than the analyzed impacts on each beach.

2. Materials and Methods

2.1 Study sites

Seven beaches from the shoreline of Rio de Janeiro state were sampled in this study. The beach choice was based on different levels of human density and their frequency of use. Most beaches are located at the urban perimeter of Rio de Janeiro city, but they still have differences in types of impact and their accessibility by the public.

(1) Barra da Tijuca - BT (23°01'S, 43°32'W) and (2) Copacabana - CP (22°97'S, 43°18'W) beaches are two of the most popular beaches in Rio de Janeiro, both located close to some of the most populous neighborhoods of the city. These beaches are constantly used for recreational purposes and are affected by the high number of visitors during the whole year, which is intensified during summer break due the high number of tourists. (3) Restinga da Marambaia - RM (23°03'S, 43°34'W) and (4) Fora - Urca - FU (22°94S, 43°15W) are also located on the urban area of Rio de Janeiro city, but both beaches are located on military area and closed for general admission. Their restricted access makes them less impacted by human density, since they are rarely used for recreational purposes and are mostly empty during the entire year. (5) Botafogo - BO beach (22°94S, 43°18W), despite also being located around populous and touristic neighborhoods of Rio de Janeiro city, does not represent an attractive environment for the public, because of the high levels of organic enrichment occurring on this beach as consequence of chronic sewage disposal (CHALEGRE TOUCEIRA et al., 2018; FISTAROL et al., 2015); therefore, it has little impact caused by human density. (6) Fora - Paraty - FP (23°34'S, 44°72'W) and (7) Guaxindiba - GX (21°47'S, 41°05'W), in São Francisco do Itabapoana, are located at the

extreme south and north of the state, respectively. These are less urbanized beaches that are mainly used during summer break and rarely used during other parts of the year. (Fig. 1)

All these beaches are classified as microtidal beaches with low declivity; beaches BT, CP, RM, FP and GX are classified as exposed and located at the oceanic portion of Rio de Janeiro state, whereas beach FU is classified as semi-exposed and beach BO as protected and both are located at Guanabara Bay. (Appendix. A)

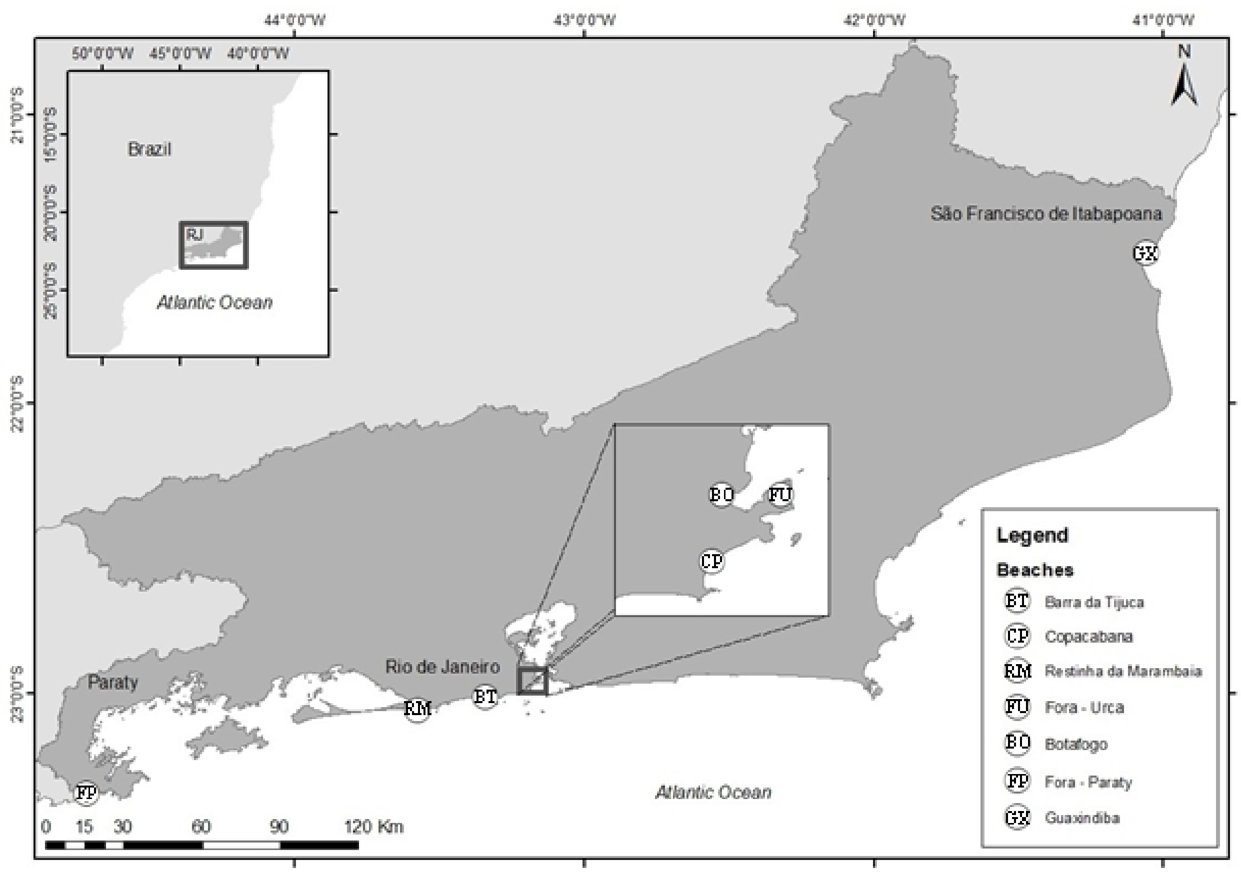


Figure 1 - Location of sampled beaches along the coast of Rio de Janeiro state.

Human density was estimated by the analysis of satellite images obtained by Google Earth between 2010 and 2016 for each one of the studied beaches. Only summer months had their images analyzed since the number of images available to be analyzed were concentrated during this period in all beaches. Human density was calculated as the average human abundance found on a standardized extension of the beach arch, close to sampling sites, in relation to the total area of each beach which was also estimated through the software. Human density was expressed by the number of individuals/100m².

2.2 Sampling strategy

Sampling occurred in March, 2015, during the spring low-tide. On each beach, a georeferenced fixed point was established at the beginning of the costal vegetation or an already existing physical structure (roads, sidewalks, walls, etc) that was used for monitoring the spring high-tide line during the sampling period. Three fixed transects equidistant by 60m from each other were established on the central portion of the beach arc. On each transect, 10 equidistant sampling stations were placed on the intertidal; station 10 represented the upper limit of the intertidal zone, but station 1 was located at the sublittoral. Distance between stations were the same for the three transects established on each beach. A total of 30 sampling stations on each beach were sampled (Fig. 2). At each station, one sediment sample was collected for meiofauna analysis, using a plexiglass core with 10cm² area introduced in the sediment to a depth of 10cm. Samples were fixed with 4% formaldehyde buffered with borax on 9:1 proportion. Two other sediment samples were collected at each station for granulometric and total organic matter analysis, placed in insulated boxes until laboratory arrival, where they were placed on a freezer (-20ºC).

YSI 6920 multiparametric probe was used to measure the main physicochemical characteristics (temperature, chlorophyll *a*, dissolved oxygen and salinity) of the water on each beach. A part of it, 100mL of water and 50g of humid superficial sediment were also sampled on each beach and placed in sterile bottles for thermotolerant coliform quantification.

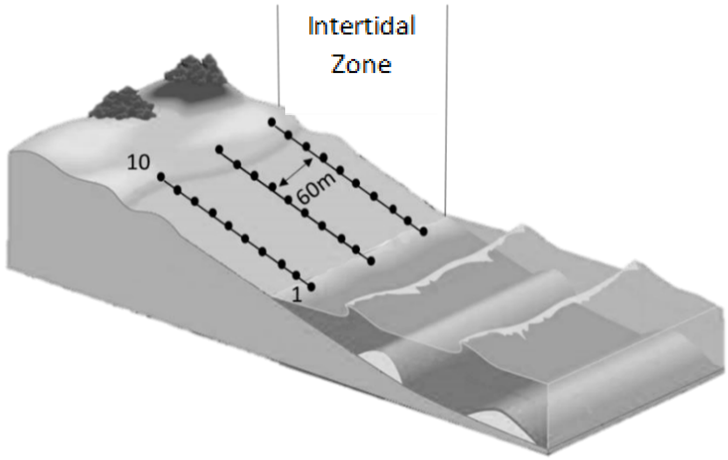


Figure 2 – Schematic illustration of the sampling strategy applied at the seven sampled sites. Numbers represent sampling sites (1: sublittoral and 10: upper limit of the intertidal zone). (Adapted from MARIA et al., 2015).

2.3 Sample Processing

For granulometry analysis, sediment samples were initially dried at 70º C until reaching constant weight. After stabilizing, samples were sieved to separate shell fragments and then grains smaller than 1mm were weighted. These fractions were later analyzed through the particle size analyzer Masterizer 2000, which determined the particle size distribution per sample. Sediment grains were classified according to the Wentworth scale (WENTWORTH, 1922) and results were presented as average for beach.

Total organic matter (TOM) was measured by a modified version of the loss on ignition technique (GREISER & FAUBEL, 1988). The excess of carbonate was removed by sieving the sediment samples to separate large shells fragments. The sediment was then oven dried at 60º C for 24h and weighted before being transferred to a muffle furnace at 450º C for 4h. Weight differences from before and after the burning process indicate the total organic matter content on each sample. %TOM per beach were showed as average.

Thermotolerant coliforms were measured by the multiple tube fermentation technique, which is the standard method for the examination of water and wastewater recommended by the *American Public Health Association* (2012). This analysis was conducted by the Water Microbiology Laboratory from the department of Environmental Sciences of UNIRIO.

Meiofauna samples were washed over a 38μm sieve, and organisms were extracted by decantation using silica at specific gravity of 1.18 (GIERE, 2009). After counting the meiofauna, 120 nematodes were randomly picked from each sample and cleared following the methodology of DE GRISSE (1969). Nematodes were mounted on permanent slides and identified on a stereoscope microscope until genus level, using pictorial keys from PLATT & WARWICK (1983, 1988) and WARWICK et al. (1998).

2.4 Biological Indexes

Taxonomic diversity, per sample, was estimated by nematode genera density data (ind.10 cm^{-2}), which was used to calculate diversity indexes as the average number of genera per sample (S), Shannon index (H'), Pielou's evenness (J') and rarefaction index $ES_{(x)}$ (expected number of genera). This last index gives an estimated number of genera present in a population of x individuals; given that the sampling site with the lowest number of nematodes sampled was 20, it was calculated for a hypothetical sample of 20 individuals ($ES_{(20)}$). These diversity indexes were calculated using software PRIMER + PERMANOVA version 6.

Functional diversity was evaluated by means of trophic diversity and maturity indexes. Feeding types followed Wieser (1953) classification. Each genus was classified as one of four feeding categories: 1A - selective deposit feeders, 1B - non-selective deposit feeders, 2A - epigrowth feeders and 2B - predators/omnivores. This classification was used to calculate the Index of Trophic Diversity (ITD) (HEIP et al., 1985), where ITD = $\Sigma\theta^2$ (θ is the percentage contribution of each feeding type according to Wieser (1953)). ITD values range from 0.25 (highest trophic diversity; i.e. the four trophic groups account for 25% each) to 1.0 (lowest trophic diversity; i.e. one feeding type accounts for 100% of total nematode assemblage).

The Maturity Index (MI) (BONGERS, 1990) was calculated as the weighted average of the individual colonizer-persistent (c-p) values. The contribution of each life-history group (c-p: 1–5) to the total nematode assemblage was used in the calculation of the MI for each beach.

All biological indexes were calculated per sample, but results were presented as an average for each beach.

2.5 Statistical Analysis

Differences in density, taxonomic and functional diversity indexes and granulometry were tested by a two-way PERMANOVA with fixed factor beach and random factor station nested on beach; total organic matter was tested by a one-way PERMANOVA for fixed factor beach. Where significant differences were found, a pairwise analysis was performed to check where those differences occurred. Before applying the PERMANOVA analysis, homogeneity of multivariate dispersion was tested using the distance among centroids by PERMDISP. K-dominance curve was used to represent the cumulative dominance of genera on each beach expressing the beta diversity. Nematode assemblage structure was analyzed by a non-metric MultiDimensional Scaling (nMDS) using Bray-Curtis Similarity on transformed density data (square root) from each sample; output was portrayed by differences among beaches and between lower (1 to 5) and upper (6 to 10) stations. SIMPER analysis was used to identify the groups that most contributed to the similarity among beaches.

A Distance Based Linear Model (DistLM) was applied to test the relationship between physicochemical and granulometric characteristics with the nematode composition of all beaches combined and of each beach individually. Conservation, Recreation and Urbanization indexes (MCLACHLAN et al., 2013; GONZÁLEZ, et al., 2014) were also calculated and used for this analysis (See supplementary material). Density data was transformed on a square root basis while environmental factors were transformed on a $\log_{(x+1)}$ basis. Variables that could not be measured in replicates were repeated (e.g. dissolved oxygen, temperature, CTE sediment, human density, conservation, urbanization and recreation indexes). Highly correlated variables were excluded. The environmental variable that best explained the nematode assemblages on most beaches was investigated into details to try explaining patterns of distribution of the main genera (relative abundance above 5% in at least one of the beaches) among its range. All statistical analyses were performed on PRIMER (v.6) + PERMANOVA add-on package.

3. Results

3.1 Environmental variables

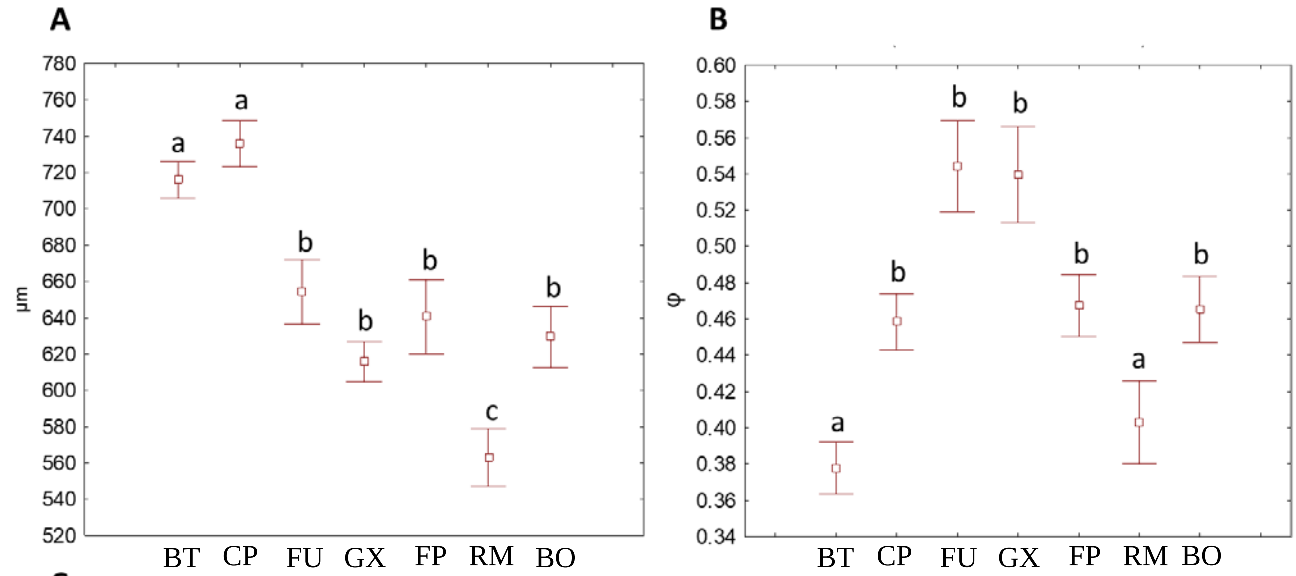
The measured physicochemical properties from the beaches showed punctual differences among them (Tab. 1). Water temperature was highest on Botafogo beach (28° C) and lowest at Restinga beach (17° C). Botafogo beach also showed the lowest values for salinity (26) and DO (4.3 mg/L) while in all other sampled beaches salinity varied from 34 to 36 and DO varied from 6.36 to 7.40mg/L. Thermotolerant coliforms were much higher on Botafogo beach, surpassing 20000 NMP/100mL in sediment samples and 30000 NMP/100mL in water samples; Fora - Paraty and Guaxindiba beaches showed values smaller than 2 NMP/100mL for the same variables. Human density peaked at Copacabana (0.76 ind./100m²) and Barra da Tijuca (0.66 ind./100m²) while Paraty and Restinga da Marambaia beaches presented null values for human density.

Table 1 - Environmental variables measured from the seven studied beaches. T: Temperature, DO: Dissolved Oxygen, TC: Thermotolerant coliforms.

	T (°C)	DO (mg/L)	Salinity	TC - water (NMP/100mL)	TC - sediment (NMP/100mL)	Human Density (100m²)
Barra da Tijuca	21.46	6.88	35	16.90	189.93	0.66
Copacabana	19.50	7.10	35	230.00	11.93	0.76
Fora - Urca	21.31	6.67	35	261.09	33.44	0.25
Fora - Paraty	26.33	6.36	34	<2.0	<2.0	0.00
Guaxindiba	25.02	6.64	34	<2.0	<2.0	0.13
Restinga da Marambaia	17.13	7.40	35	8.60	91.67	0.00
Botafogo	28.53	4.30	26	31516.67	20593.33	<0.01

Median grain size ranged from 472 to 972 µm, values that allowed the classification of all beach sediment as coarse sand. The grain size was significantly higher at Copacabana and Barra da Tijuca beaches (735±13 and 715±10µm, respectively) and smaller at Restinga da Marambaia beach (563±16 µm) (Fig. 3A). The sorting coefficient ranged from 0.31 to 0.90 φ (indicating moderately to very well sorted grains) (Fig. 3B). It was significantly smaller on Barra da Tijuca and Restinga da Marambaia beaches (0.38 and 0.40 φ, respectively) and did not statistically differ among the other beaches.

Total organic matter ranged from 0.06 to 4.91% and averages were significantly higher at Botafogo (1.15%) and Guaxindiba beaches (0.79%) (Fig. 3C). The remaining beaches showed significantly less %TOM, with values ranging from 0.11 and 0.26%.



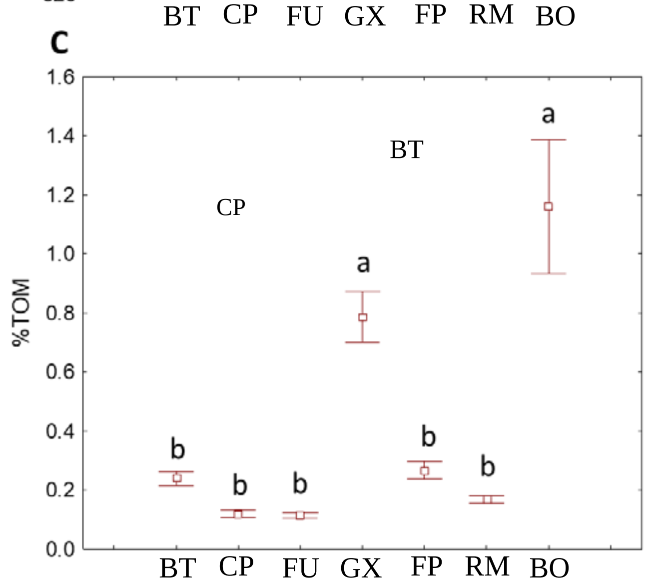


Figure 3 - Median grain size (A), sorting coefficient (B) and total organic matter (C) on the seven sampled beaches: BT = Barra da Tijuca, CP = Copacabana, FU = Fora - Urca, GX = Guaxindiba, FP = Fora - Paraty, RM = Restinga da Marambaia, BO = Botafogo. Lower case letters represent pairwise PERMANOVA results (n = 30, error bars = standard error).

3.2 Biological data

A total of 100 nematode genera from 35 different families were found on the seven sampled beaches. Xyalidae, Chromadoridae and Thoracostomopsidae were the most representative families. The smallest number of identified genera per beach was 29 on Botafogo beach, which was highly dominanted by *Chromadorita* (Tab. 2), and the highest was 53 on Restinga da Marambaia beach, where *Paracanthonchus* and *Daptonema* accounted for more than 50% of the identified organisms. At Copacabana and Fora - Urca beaches 43 and 36 genera were found, respectively, both showing high relative abundance of *Daptonema*, *Theristus* and *Microlaimus.* Meanwhile Guaxindiba had 40 identified genera, with *Theristus, Latronema* and *Daptonema* as the most representative organisms. *Ascolaimus* was the second most frequent genus at Fora - Paraty and Barra da Tijuca beaches, where 37 and 48 genera were found, respectively. In these latter beaches, *Epacanthion* and *Leptolaimus* had the highest relative abundance, respectively. (Tab. 2).

Nematode density ranged from 2 to 6704 ind.10cm^{-2} on all beaches and presented significantly differences among beaches. Highest significant values were found on Fora - Urca beach (1314±226 ind.10cm^{-2}) and smallest on Fora - Paraty and Barra da Tijuca beaches (40±8 and 73±9 ind.10cm^{-2}, respectively) (Fig. 4A).

Generic richness (S) (Fig. 4B) had a maximum average value at Guaxindiba beach (9.9±0.4) and minimum at Botafogo beach (6.2±0.4) and Shannon diversity (H') varied from 1.01±0.1 at Botafogo beach to 1.49±0.1 at Barra da Tijuca beach (Fig. 4C). Although PERMANOVA showed significant differences among beaches (Appendix. B), the pairwise analysis was not able to detect where those differences occurred. Equitability (J) was significantly higher on Barra da Tijuca and Fora - Paraty beaches, both with values above 0.75 (Fig. 4D). $ES_{(20)}$ had smaller values on Botafogo beach, but significant differences among beaches were not found (Fig. 4E).

ITD varied from 0.45 at Barra da Tijuca beach to 0.69 at Botafogo beach, but only Barra da Tijuca showed significantly smaller values for ITD when compared to the other six sampled beaches (Fig. 4F). The MI had similar values for all beaches and did not significantly differ among beaches, varying from 2.38 (Botafogo beach) to 2.61 (Fora - Paraty beach) (Fig. 4G).

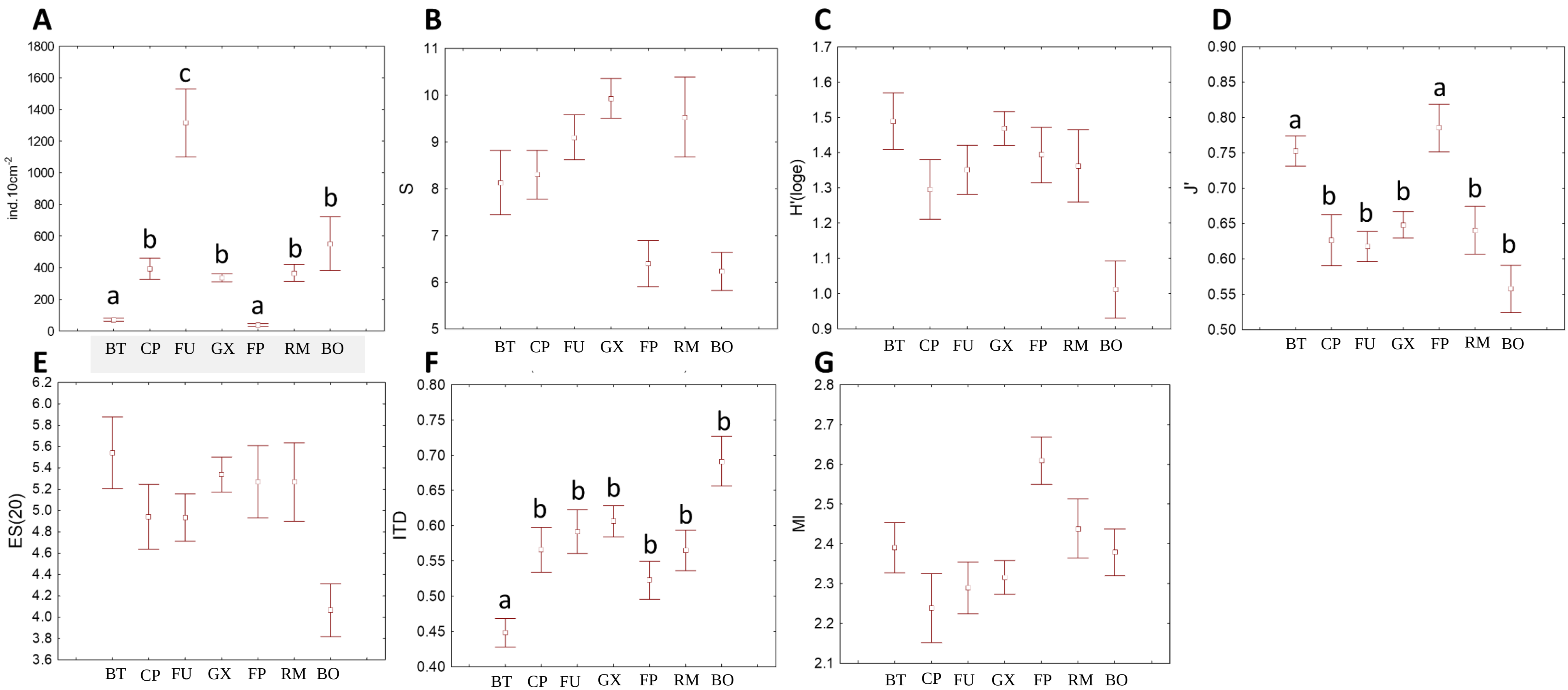


Figure 4 - Biological indexes calculated for each beach: Density (A), Generic richness (B), Shannon diversity (C), Equitability (D), $ES_{(20)}$ (E), ITD (F) and MI (G). BT = Barra da Tijuca, CP = Copacabana, FU = Fora - Urca, GX = Guaxindiba, FP = Fora - Paraty, RM = Restinga da Marambaia, BO = Botafogo. Lower case letters represent pairwise results of the indexes with significant differences; graphics without letters represent indexes without significant differences (n = 30, error bars = standard error).

Table 2 - Relative abundance (%) of the main genera found on the study sites. Table presents genera with relative abundance higher than 5% on at least one sampled beach; Others are the sum of the remaining genera for each beach.

Genera	Beaches						
	Barra da Tijuca	Copacabana	Fora - Urca	Guaxindiba	Fora -Paraty	Restinga da Marambaia	Botafogo
Ascolaimus	12.67	5.19	4.76	7.20	13.61	2.81	0.16
Chromadorita	4.33	11.84	5.64	0.22	5.92	0.00	58.41
Daptonema	4.60	27.13	20.62	14.23	2.13	21.21	2.02
Enoplolaimus	0.98	1.00	5.47	0.00	0.08	0.00	0.00
Epacanthion	0.65	1.09	0.53	5.89	31.20	0.44	0.00
Latronema	0.00	0.00	0.18	17.92	0.15	0.00	0.00
Lauratonema	0.11	0.00	10.36	0.00	0.00	2.12	0.00
Leptolaimus	18.97	0.32	0.10	0.71	2.60	0.02	0.00
Marylynnia	7.03	7.22	1.38	0.00	0.00	1.53	0.04
Metachromadora	9.89	0.10	0.32	8.06	0.00	2.68	10.19
Microlaimus	1.14	13.97	19.83	2.76	1.04	0.08	0.01
Odontophora	0.05	0.00	0.60	0.52	0.00	0.19	6.06
Oncholaimus	3.24	2.27	0.00	0.00	10.00	0.05	0.72
Paracanthonchus	0.12	0.00	0.00	0.67	1.99	30.75	0.03
Paracyatholaimus	0.00	0.19	0.00	1.08	0.00	6.49	0.00
Pseudosteineria	2.45	2.29	0.90	4.65	7.13	0.14	0.01
Southerniella	0.00	0.00	0.00	0.00	0.00	0.00	10.27
Thalassomonhystera	7.18	3.11	0.44	0.00	2.29	0.00	0.00
Theristus	4.75	14.92	18.79	22.52	3.08	8.05	4.16
Trissonchulus	7.81	0.76	0.79	0.06	0.09	4.39	0.03
Others (n)	14.03 (31)	8.60 (28)	9.29 (20)	13.51 (26)	18.69 (23)	19.05 (38)	7.89 (16)

K-dominance curve (Fig. 5) showed Botafogo as the least diverse beach, whereas one single genus (*Chromadorita*) accounted for almost 60% of the relative abundance of nematodes on this beach. Barra da Tijuca beach had the lowest cumulative dominance, with *Leptolaimus* (19%), *Ascolaimus* (13%) and *Metachromadora* (10%) as the most representative genera on this beach (Tab. 2).

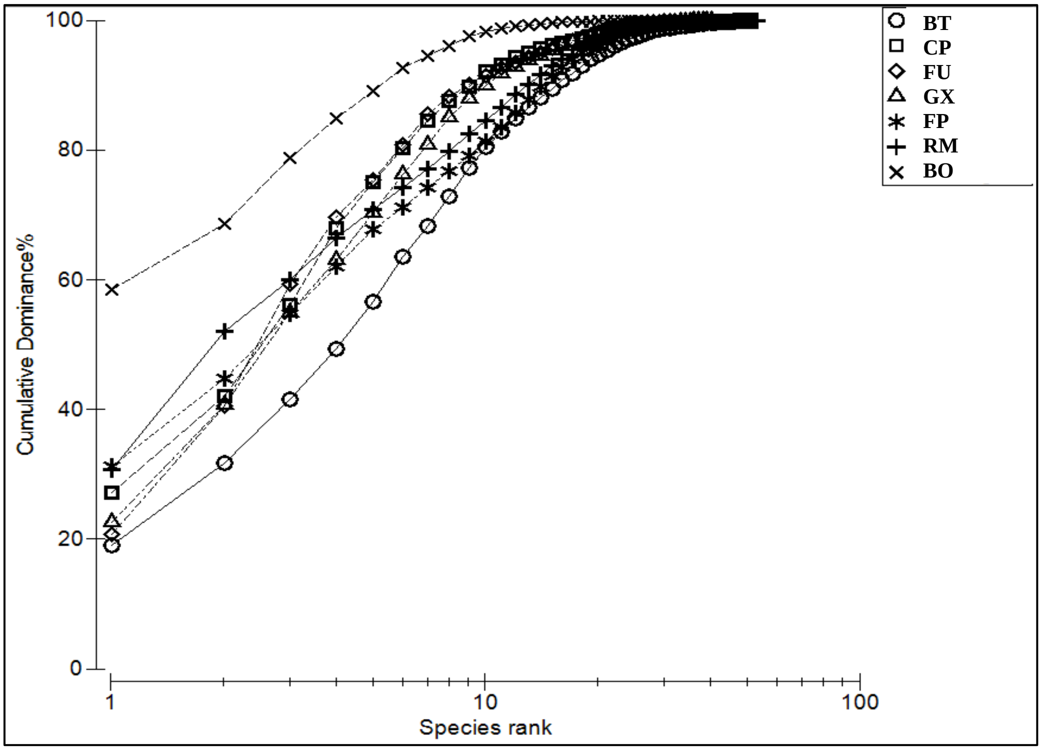


Figure 5 - K-dominance curves for nematode genera on each of the seven sampled beaches: BT = Barra da Tijuca, CP = Copacabana, FU = Fora - Urca, GX = Guaxindiba, FP = Fora - Paraty, RM = Restinga da Marambaia, BO = Botafogo.

The non-metric MDS (Fig. 6) showed that nematode assemblages from Botafogo and Paraty beaches formed two different groups, which can be easily distinct from the nematode assemblages of the other five beaches (Fig. 6A). Pairwise analyzis for the assemblage showed significant differences between all analyzes beaches. It was also observed a clear separation between upper beach (stations 6 to 10) and lower beach/sublittoral (stations 1 to 5) (Fig. 6B). SIMPER analysis (Appendix. C) showed that *Daptonema* and *Theristus* contributed most for the similarity of Copacabana, Fora - Urca and Guaxindiba beaches, whereas the other four beaches had *Leptolaimus Ascolaimus*, *Paracanthonchus* and *Odontophora* contributing to the similarity of Barra da Tijuca, Fora - Paraty, Restinga da Marambaia and Botafogo, respectively.

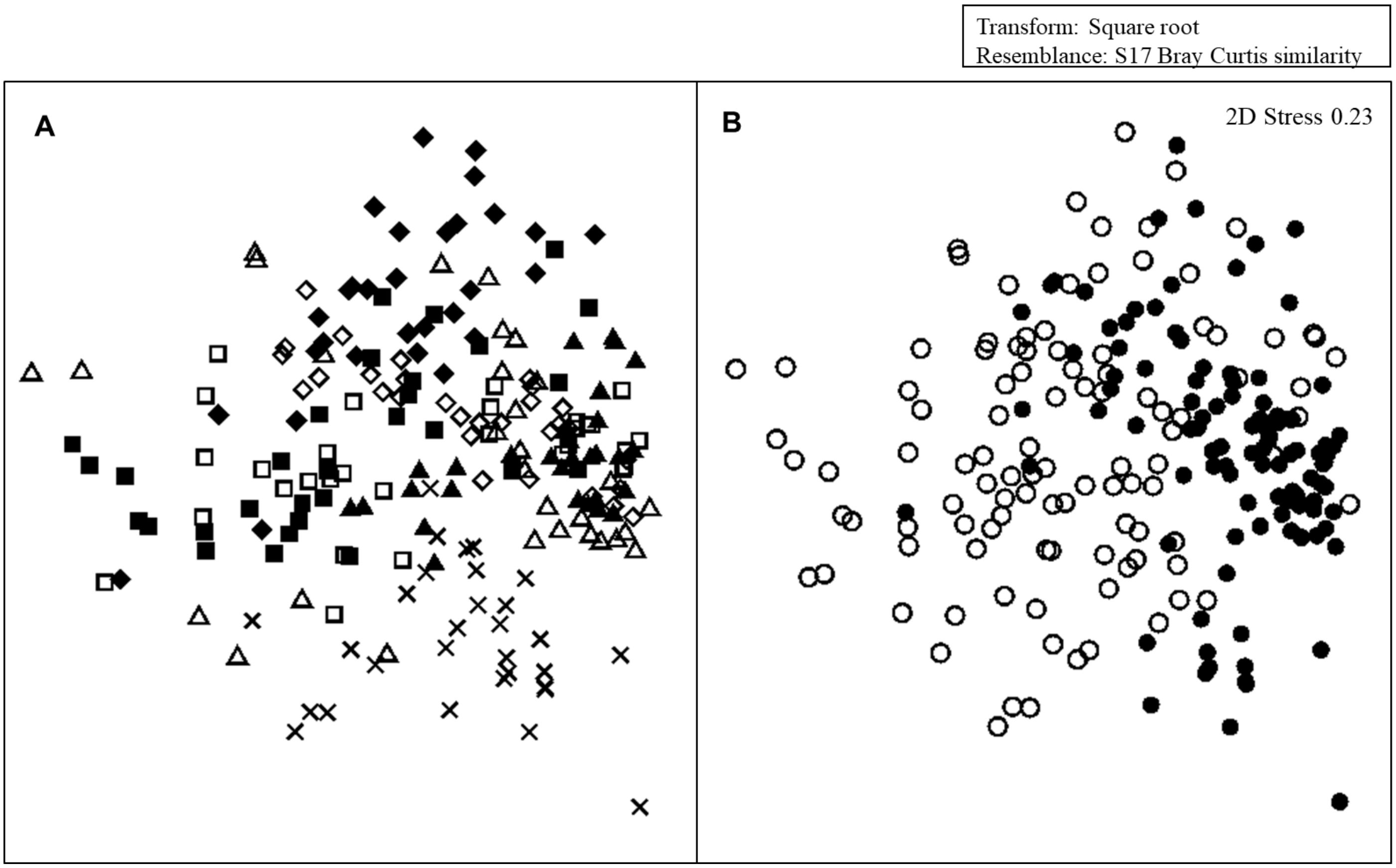


Figure 6 - Output of non-metric MultiDimensional Scaling (MDS) based on the transformed nematode assemblages similarity for different beaches (A) and different stations (B). 6A: ■ = Barra da Tijuca, □ = Copacabana ▲ = Fora - Urca, Δ = Restinga da Marambaia, ◊ = Guaxindiba, ♦ = Fora - Paraty and **x** = Botafogo. The same simbols were used for beaches with similar gradients of impact. 6B: ● = lower beach/sublittoral stations, ○ = upper beach stations.

3.3 Relationship between nematode assemblage structure and environmental variables

The DistLM routine showed that non-correlated measured environmental variables together were able to explain 32% of the nematode assemblage structure of all the beaches together (Tab. 3). In this case, dissolved oxygen and the sorting coefficient were the two variables that best explained the variability of the nematode assemblage from all the beaches together. For the DistLM routine applied for each beach individually, the sorting coefficient was the most frequent variable that best explained nematode assemblage of Copacabana (13%), Fora - Urca (24%), Guaxindiba (21%), Fora - Paraty (8%) and Restinga da Marambaia (10%) (Tab. 3). TOM also had significant result for most beaches and best explained the distribution of Barra da Tijuca beach (22%). Human density only significantly explained the nematode assemblage of Copacabana beach (8%). None of the variables explained the nematode assemblage of Botafogo beach.

Table 3 - Results of the DistLM analysis for the whole nematode assemblage and for each sampled beach separately. *Prop* is the proportion of the variability explained; *Cumul is the* cumulative proportion of the variability explained.

		AIC	SS(trace)	Pseudo-F	P	Prop.	Cumul.
Nematode Assemblage (including all 7 beaches)	DO	1710.4	53579.0	15.70	0.0001	0.07	0.07
	Sorting Coefficient	1699.3	42905.0	13.31	0.0001	0.06	0.13
	Temperature	1688.8	38658.0	12.67	0.0001	0.05	0.18
	Conservation Index	1680.7	29544.0	10.11	0.0001	0.04	0.22
	Urbanization Index	1673.8	24766.0	8.80	0.0001	0.03	0.25
	Human Density	1666.3	25234.0	9.33	0.0001	0.03	0.28
	Mean Grain Size	1663.5	12441.0	4.68	0.0001	0.02	0.30
	Recreation Index	1662.4	7889.0	3.00	0.0003	0.01	0.31
	CTE Sediment	1662.1	5733.0	2.19	0.0058	0.01	0.31
	TOM	1661.9	5027.0	1.92	0.0192	0.01	0.32
Barra da Tijuca	TOM	237.1	19588.0	7.73	0.0001	0.22	0.22
Copacabana	Sorting Coefficient	240.6	11744.0	4.11	0.0028	0.13	0.13
	Human Density	239.8	7252.1	2.69	0.0100	0.08	0.21
Fora - Urca	Sorting Coefficient	223.1	13862.0	8.72	0.0001	0.24	0.24
	TOM	221.5	4993.6	3.41	0.0029	0.09	0.32
	Mean Grain Size	220.7	3536.4	2.56	0.0232	0.06	0.38
Guaxindiba	Sorting Coefficient	228.2	13892.0	7.36	0.0010	0.21	0.21
	TOM	226.7	5951.4	3.43	0.0132	0.09	0.30
Fora - Paraty	Sorting Coefficient	241.1	7272.6	2.51	0.0054	0.08	0.08
	TOM	240.3	7216.1	2.63	0.0055	0.08	0.16
Restinga da Marambaia	Sorting Coefficient	242.9	9153.4	2.98	0.0070	0.10	0.10
Botafogo	Human Density	237.4	0	0	1	0	0

3.4 Nematode assemblage and Sorting Coefficient

Nematode distribution was analyzed through the range of sorting coefficient found on each sample from each beach (Fig. 7). Some genera seem to be linked to some sorting spectrums. For instance, *Daptonema* and *Theristus* were found in high relative abundance on a broad spectrum of the sorting coefficient (0.3 - 0.9 φ, ranging from moderately sorted to very well sorted), whereas other genera had their distribution associated to a smaller range of the sorting coefficient values: *Ascolaimus, Chomadorita* and *Odontophora* showed association with all sorting coefficients ranging from 0.3 to 0.55 φ (very well sorted to moderately well sorted); *Metachromadora* and *Microlaimus* were more associated with spectrums 0.5 and 0.55 φ (moderately well sorted); *Latronema* and *Leptolaimus* showed connection with ranges from 0.3 to 0.4 φ (very well sorted to well sorted); *Paracanthonchus*, *Thalassomonhystera* and *Trissonchulus* were mainly found on sorting coefficients around 0.3 to 0.35 φ (very well sorted) and *Epacanthion* showed higher connection with spectrums around 0.45 and 0.5 φ (well sorted). The other seven analyzed genera (*Enoplolaimus, Lauratonema, Marylynnia, Oncholaimus, Paracyatholaimus, Pseudosteneria and Southerniella*) did not show a clear association of their relative abundance to any value of the sorting coefficient spectrum.

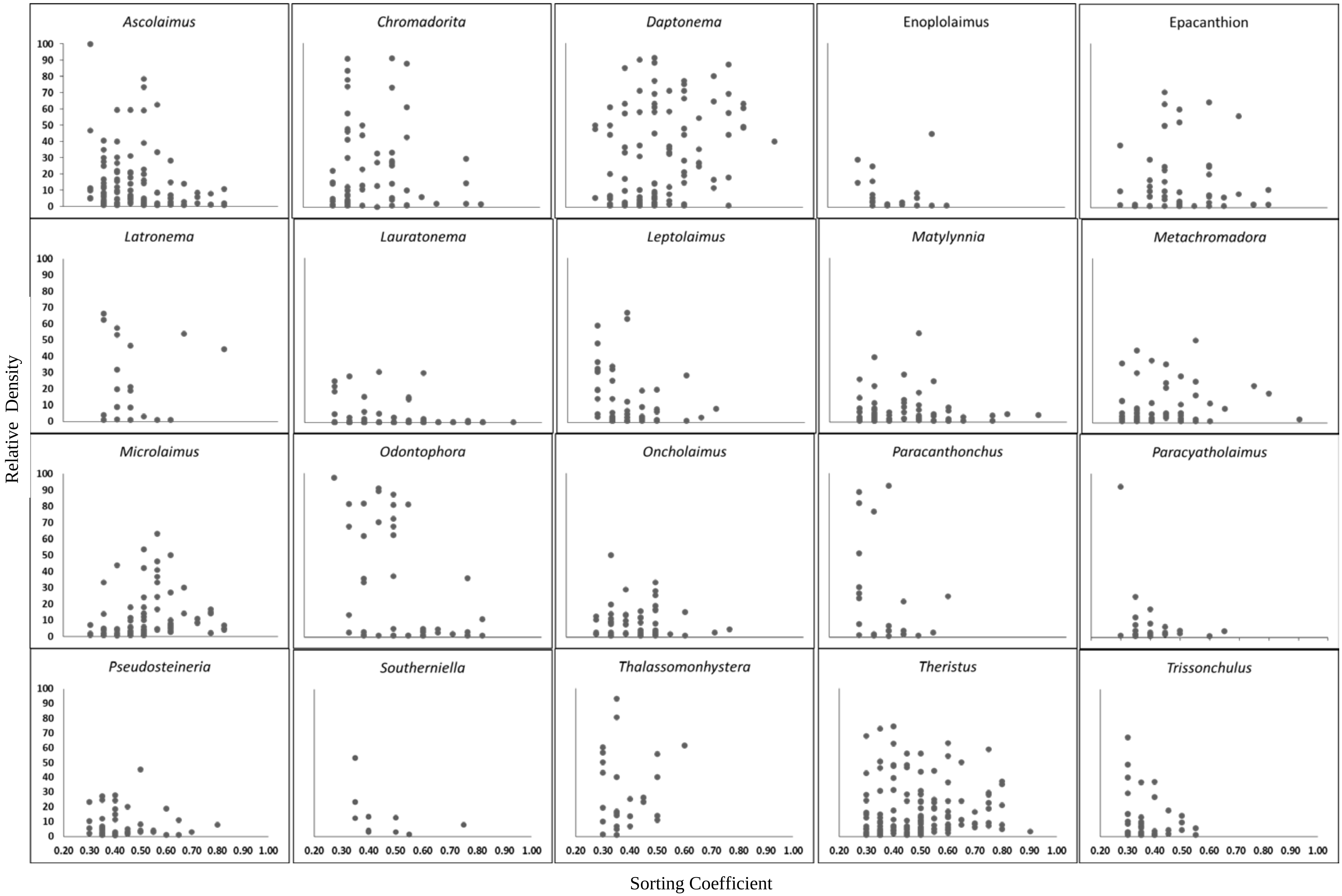


Figure 7 - Distribution of the main nematode genera according to the sorting coefficient spectrum analyzed for all seven beaches grouped. Data was standardized for each genus using the maximum density as 100% of relative abundance.

4. Discussion

Given the high importance of sandy beaches for coastal cities on tropical countries and the growth of biomonitoring as a reliable technique of ecological assessment, this study aimed on trying to evaluate the ecological *status* and characterize seven different sandy beaches from the state of Rio de Janeiro based in their nematode assemblages and their relationships with the measured environmental variables. Our results indicated that physical variables such as the sorting coefficient might represent a more important role of nematode distribution on sandy beaches when compared to anthropic impacts that were measured in this research; some nematode genera seem to have a close relationship with specific spectrums of sorting due to their autoecology. The seven sampled beaches present many differences concerning the type of anthropic disturbance that they are exposed to and we examined if nematode assemblage is directly affected by these impacts. Whereas Botafogo beach is considered the most degraded beach due to the chronical sewage disposal, the remaining studied beaches are mostly affected by tourism and recreational activities, especially Copacabana and Barra da Tijuca beaches, where these activities are almost never uninterrupted during the whole year followed by accentuated use during summer.

The least diverse nematode assemblage was found in the most degraded beach (Botafogo) showing the most isolated nematode assemblage in the nMDS. These results could reflect the distinct values found for most environmental measured variables, such as temperature, dissolved oxygen, salinity, thermotolerant coliforms and, mainly TOM, when compared to the other beaches. For instance, the last environmental variable had much higher values than those found for the other beaches and reflected the organic enrichment, which is known to be the highest impact on the ecological *status* of Botafogo beach (CHALEGRE TOUCEIRA et al., 2018). Low values for salinity and dissolved oxygen have also been recorded on other Guanabara Bbay characterization studies and is also associated with the sewage input that has been impacting Botafogo beach for decades (FISTAROL et al., 2015, PARANHOS & MAYR, 1993). On this beach, the highest dominance of the genus *Chromadorita* played an important role on the isolation of its nematode assemblage contradicting a previous study from a temperate beach, which revealed that this genus was associated to pristine conditions (SEMPRUCCI et al., 2015). Since *Chromadorita* is classified as an epigrowth feeder, benthic microalgae may represent an important food source for these organisms (MOENS & VINCX, 1997). Unfortunately, the availability of chlorophyll *a* in the sediment (a proxy for benthic macroalgae) was not measured in this study. It is also not even mentioned as an important variable on the standard guidelines from the Brazilian Protocol for Monitoring Benthic Coastal Habitats (TURRA & DENADAI, 2015). However, this variable had already been measured on other sandy beaches ecological studies (e.g. MORENO et al., 2008; MARIA et al., 2012) and presented positive links with meiofauna distribution (SUN et al., 2014) revealing their necessity of being measured in future biomonitoring studies.

On the other hand, despite the broad recreational utilization and higher values of human density found in Copacabana and Barra da Tijuca beaches, values for most applied biological indexes (S, H', $ES_{(20),}$ MI) did not significantly differ from the remaining sampled beaches, which are less affected by human presence. Both beaches are located far from common sources of chemical impact such as rivers and harbor activities and are constantly being cleaned up by city management, which also reduces damages caused by the large waste disposal made by visitors (DE CARVALHO & BAPTISTA NETO, 2016). Apart of it, Barra da Tijuca had the highest equitability (alongside with Fora-Paraty beach), lowest cumulative dominance and the lowest value for ITD, i. e. presenting highest beta and trophic diversity. This beach shows a large spatial difference according to levels of anthropogenic pressures on its beach arc (VELOSO et al., 2006); some portions, such as our selected sampling site, are located further from central urban areas, transportation is less available, and construction is prohibited (CARDOSO et al., 2016). Although these conditions do not reflect the majority of the beach arc, it may indicate that the lack of higher urban impacts may be favoring a better ecological *status* of some portions of Barra da Tijuca beach as it was recorded previously on macrofaunal studies performed on the same beach (e.g. VELOSO et al., 2006; CARDOSO et al., 2016).

Concerning the nematode density, it varied according previous studies on Brazilian sandy beaches and other tropical beaches with similar granulometric properties (e.g. VENEKEY et al.,2014; MARIA et al.; 2012, KOTWICKI et al., 2005). Differences in nematode density are usually related to the median grain size present on each beach, in general coarse sediment favors higher nematode densities (GIERE, 2009). Since all sampled beaches were classified as having coarse sand we expected to find a small density variability; however, the large range of nematode density among beaches (40±8 ind.10cm^{-2} in Barra da Tijuca to 1314±226 ind.10cm^{-2} in Fora - Urca) could be associated to a non-measured environmental variable, such as precipitation (GIERE. 2009) as sampling occurred during the rainy season.

The formation of two different groups portrayed by the nMDS analysis showed the separation of the lower sampled stations from the upper stations. The upper stations (slightly smaller grains/ better sorted) showed higher differences in assemblage distribution among beaches when compared to the lower stations, that showed more overlaps and similarities among them. Upper beach stations have higher influence of physical factors, such as desiccation and anthropic impacts including trampling and microplastic/waste disposal (KNOX, 2001). Since these impacts occur in different extents among beaches, it would allow the more adapted genus to enhance its densities on each beach, as it was found that abundance of most dominant genera on each beach was higher on upper stations of the sampling sites. In contrast, the overlaps of distribution on the lower stations (slightly coarser grains/ more poorly sorted) could be a result of the amelioration of the extreme conditions faced by upper stations. Additionally, as the lower stations also remain submerged for most part of a tidal cycle, nematode assemblage renewal is expected to happen more frequently as many organisms may be brought to the sediment by the water movement (MCLACHLAN, 1980).

Altogether, the applied biological indexes were not able to present completely clear answers concerning the ecological *status* of the seven studied beaches as most values of diversity (i.e. S, H',J and $ES_{(20)}$) and functional (i.e. ITD, MI) measurements were similar among beaches and could not directly explain the nematode assemblage distribution. DistLM analysis showed that the sorting coefficient played the most important role on the nematode assemblage distribution for the seven sandy beaches. As the morphodynamic state of the beach is composed by a large group of interacting variables (i.e. slope, porosity, wave characteristics), it is difficult to isolate effects of each one of them on assemblage composition (RODRIGUEZ, 2004), especially when the differences are not clearly affecting sediment properties as it was the case of all seven sampled beaches from this study since they are exposed (Barra da Tijuca, Copacabana, Fora - Paraty, Restinga da Marambaia, Guaxindiba), semi-exposed (Fora - Urca) or protected (Botafogo). However, the relationship of nematodes with the mean grain size has been widely investigated, and although it is accepted that coarser sediments bear high species richness and diversity than finer sediments (HEIP, 1992), it is debated that this single variable cannot solely explain nematode diversity and density on sandy beaches (FONSECA et al., 2014; SNELGROVE & BUTMAN, 1994). It is postulated that grain size also affects assemblage composition, as coarser sediments favors dominance of families Xyalidae, Cyatholaimidae, and

Chromadoridae as found in this study and finer sediments are characterized by families Desmodoridae and Linhomoeidae (HEIP et al., 1985; GHESKIERE et al., 2005). As sandy beaches will never be composed of uniform sediment particles, their sediment characterization also relies on other granulometric properties, like sorting coefficient, skewness, and kurtosis (MCLACHLAN & TURNER, 1994). Consequently, these variables may also display important ecological influence on the distribution of interstitial organisms as nematodes. Their assemblages may be responding to them instead of the mean grain size itself. Nowadays, there are gaps of information concerning to the influence of those granulometic properties, especially the sorting coefficient, on nematode assemblage distribution and how it may affect diversity.

In our study, the sorting coefficient better explains the nematode assemblage found in the seven sandy beaches. The sorting coefficient reflects the severity of the hydrodynamic regime on the sediment, since grains are mainly shaped by tidal movements and the action of waves (MCLACHLAN & TURNER, 1994; URBAN-MALINGA et al., 2004), and influences sediment characteristics like porosity and permeability. More poorly sorted sediments are known to offer more spaces and niches for nematodes to colonize when compared to well sorted sediments that usually gives narrower niches for meiofaunal organisms (URBAN-MALINGA et al., 2004; COLEMAN et al., 1997; MARCOTTE, 1986). In contrast, poorly sorted sediments may also be a limiting variable for some nematode genera as it offers less pore volume available for intertidal organisms and may favor genera with slender body shapes (SNELGROVE & BUTMAN, 1994). For instance, predators with bigger body volumes like *Trissonchulus*, *Epacanthion* and *Metachromadora* seems to be benefited from smaller sorting values since their occurrence is associated to very well sorted to moderately well sorted sediments (0.3 to 0.55 φ) in our study. Opportunistic genera with smaller body sizes such as *Daptonema* and *Theristus* seem to be able to thrive on a broader range of sorting values, as their trophic behavior of non-selective deposit feeders enables them to survive on a large variety of environments (WILSON & KHAKOULI-DUARTE, 2009; MARTINS et al., 2015; BONGERS & FERRIS, 1991). Adaptations that facilitate locomotion between grains, like caudal adhesive glands present on some *Theristus* species (ADAMS & TYLER, 1980), are also morphological features that may influence on the range of sorting that this genus can colonize.

5. Conclusion

As results showed that the sorting coefficient represented the main variable responsible for nematode distribution, this study is the first attempt to associate the grain sorting with some nematode genera, showing that some of them appears to be linked with specific spectrums of this variable. Although the applied biological indexes were not able to explain the different impacts occurring in each studied beach, it was possible to assess that these beaches have dissimilar nematode composition and that grain size may not always be the primary factor responsible for nematode distribution on sandy beaches. This is also the first study that tried to compare nematode assemblage distribution with the ecological *status* of sandy beaches from Rio de Janeiro, including the first nematode assessment for most of them, which represents an important baseline for further monitoring projects.

6. Acknowledgements

This article was part of the project “Impactos antrópicos em praias fluminenses: uma abordagem ecológica utilizando bioindicadores naturais” that was financially supported by FAPERJ – E-26/010.001508/2014. It was also financed in part by the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior - Brasil (CAPES) - Finance Code 001. We would like to acknowledge Guilherme Maricato for creating the used maps, Tatiana Cabrini for providing the morphodynamic information of the studied sites, Ana Clara Rocha for helping with the human density calculation, Ricardo Polery for allowing us to process granulometry samples on the Laboratory of Biogeochemistry-UFRJ and Haelly Dragnev for providing graphical technical support. Raíssa Vieira e Luis Otávio de Lima are also acknowledged for all the support with sampling, nematode extraction and partial nematode identification.

We acknowledge the three anonymous reviewers for the helpful insights and Sean A. Watkins for kindly proofreading this manuscript.

Declarations of interest: none

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Apêndices do Capítulo I

Appendix A - Morphodynamic characteristics of the seven studied sandy beaches (CABRINI, personal communication)

	Beaches						
	Copacabana	Barra	Restinga	Fora-Urca	Botafogo	Fora-Paraty	Guaxindiba
Surf zone (m)	50	20	25	8	50	5	20
Wave height (m)	1.00	0.50	0.30	0.30	-	0.40	0.20
Wave period (s)	76.27	96.50	94.85	80.50	-	88.31	94.33
Swash width (m)	14	9	15	16	46	9	13
Swash time (s)	65.35	57.32	53.09	62.41	1.21	52.84	120.00
Slope	0.07	0.16	0.05	0.09	0.06	0.08	0.09

- : Wave height and period are not presented since it is a protected beach.

Appendix B - PERMANOVA results for environmental variables, nematode density and biological indexes applied on the studied beaches.

Variables	Beach				Station(beach)			
	df	MS	Pseudo-F	P	df	MS	Pseudo-F	P
Mean grain size	6	111320	8.370	**0.001**	63	13147	2.487	**0.001**
Sorting coefficient	6	0.141	5.969	**0.001**	63	0.023	2.688	**0.001**
TOM	6	1,64	5.241	**0.001**	-	-	-	-
Nematode Density	6	29495	12.940	**0.001**	63	2279	2.399	**0.001**
S	6	63.533	3.121	**0.010**	63	20.355	3.064	**0.001**
H'	6	0.760	2.233	**0.049**	63	0.340	2.243	**0.001**
J	6	0.191	5.406	**0.001**	63	0.035	1.410	**0.045**
$ES_{(20)}$	6	7.079	1.519	0.181	63	4.659	2.336	**0.001**
ITD	6	0.169	3.631	**0.005**	63	0.046	2.512	**0.001**
MI	6	204.840	1.752	0.120	63	117	3.167	**0.001**
Nematode Assemblage	6	32603	5.6297	**0.001**	63	1449.7	3.9949	**0.001**

Appendix C - Results of the SIMPER analysis showing the contribution (%) of the most relevant genera to each beach, cut-off at 50%. Overall similarity for each beach is presented in brackets.

	Barra da Tijuca (49%)	Copacabana (55%)	Fora - Urca (57%)	Guaxindiba (71%)	Fora - Paraty (34%)	Restinga da Marambaia (45%)	Botafogo (57%)
Ascolaimus	12			10	35	7	
Chromadorita							17
Daptonema		17	22	14		13	
Epacanthion					16		
Latronema				9			
Leptolaimus	20						
Microlaimus		9					
Metachromadora						6	
Odontophora							34
Paracanthonchus						15	
Thalassomonhystera	10	10					
Theristus		12	22	17		9	
Trissonchulus	11						

Annex 1 - Guidelines for Conservation, Recreation and Urbanization indexes calculation (MCLACHLAN et al., 2013; GONZÁLEZ, et al., 2014).

Index of Conservation Value (CI)

Category	Condition and Score					
Dunes	**0** Absent, replaced by hard engineering structures	**1** Severely disturbed and limited in extent	**2** Extensive disturbance	**3** Disturbed but largely intact	**4** Well developed, little disturbance	**5** Pristine and extensive
Endangered and iconic species	**0** Absent	**1** Presen in low numbers, not nesting	**2** Present in good numbers, may be nesting	**3** Nesting/spawning present in large numbers		
Macrobenthic diversity and abundance	**0** Low abundance, reflective and/or short beach	**1** Intermediate	**2** Species rich and abundant, dissipative and/or long beach			
Total score	**Minimum score is 0 + 0 + 0 = 0; Maximum score is 5 + 3 + 2 = 10**					

Index of Recreation Value (RI)

Category	Condition and Score					
Infrastructure	**0** No infrastructure, difficult access	**1** No infrastructure, limited access	**2** Modest infrastructure, reasonable access	**3** Good access, some amenities	**4** Good infrastructure and access	**5** Excellent access, parking and amenities, inclusing lifesaving
Safety and health	**0** Extremely hazardous and/or polluted	**1** Hazardous and/or polluted	**2** Moderate hazards and clean	**3** Low bathing hazards, clean and totally pollution free		
Physical carrying capacity	**0** Limited, pocket beach, no backshore	**1** Intermediate	**2** Extensive beach with wide backshore			
Total score	**Minimum score is 0 + 0 + 0 = 0; Maximum score is 5 + 3 + 2 = 10**					

Index of Urbanization Value (UI)

	Beach urbanization indicator levels		
	Low 0–1	Medium 2–3	High 4–5
Proximity to urban centers	Sector with rural character. Several kilometers away from urban center. No direct influence of an urban center on the beach.	Sector located c. 1 km from an urban center, showing some effects on the beach, such as noise, some lighting and nearby vehicles passing.	Sector just meters from an urban center, the city virtually integrated. The beach is next to vehicular traffic, with evident noise and urban lighting.
Buildings on the sand	No nearby buildings appreciable.	There are buildings close to the beach but not on the sand or the dunes.	There are buildings that occupy the space at the beach or in the dunes.
Cleaning of the beach	The beach is not ‘‘cleaned’’ by mechanical means, with no sand removal.	Although mechanically engaged for cleaning, this is done infrequently, no more than 1 time per week. No frequent removal of sand.	Beach is repeatedly cleaned by mechanical means, more than once a week, which causes frequent removal of the sand.
Solid waste on the beach	No waste in the sand or the amount of waste on the beach is minimal.	In a short walk of a few meters, some solid waste can be seen on the sand, such as paper, plastic containers and cigarette butts.	Clearly a high frequency of solid waste on the sand, including papers, plastic containers, cigarette butts, plastic debris, scrap wood and glass.
Vehicles traffic on the sand	No vehicle tracks were observed on the sand. There is no vehicle access on the beach.	Although there are traces of vehicle passage, they are scarce. Vehicular crossing is not periodic and not constant. Vehicular access to the beach is relatively limited.	There are many tracks, showing recurring vehicular traffic. Various vehicles have access to the beach.
Quality of the night sky	Sky conditions are optimal for stargazing. The sky appears black, and hundreds of stars can be seen perfectly.	The glow of artificial light moderately impaired conditions for stargazing. The sky appears dark gray, and some tens of stars can be seen with some difficulty.	Given the high brightness of the artificial lighting, the conditions for stargazing are bad. The sky is gray and occasional stars can be seen. Light pollution is evident.
Frequency of visitors	The area is visited by very few people, and those are located in areas isolated from each other. Rural beach.	The sector has a moderate demand for use. Although it has tourists, based on either location or privacy, it does not have a large number of users.	Sector in high demand from users, considered a high tourist beach. Public access urban beach.

3. CONCLUSÕES GERAIS

- Os resultados do presente trabalho indicam que o uso dos nematódeos como ferramentas de biomonitoramento devem ser analisados com cautela, uma vez que a comunidade pode estar apresentando respostas principalmente em relação às variáveis ambientais encontradas no ambiente, como o grau de seleção. É importante ressaltar também que a falta de réplicas para algumas das variáveis analisadas (índices de conservação, recreação e urbanização, coliformes termotolerantes e densidade humana) também pode alterar a percepção da verdadeira importância dos impactos em cada uma das praias na distribuição dos nematódeos.

- As análises estatísticas indicam que o grau de seleção foi a principal variável responsável pela distribuição das comunidades nematofaunísticas nas praias arenosas do estado do Rio de Janeiro, com importância superior ao tamanho médio do grão, que é tradicionalmente dita como a principal variável a influenciar na distribuição desses organismos.

- Alguns gêneros de nematódeos parecem possuir maior afinidade a determinados espectros de grau de seleção, o que provavelmente está relacionado à autoecologia de cada um dos gêneros e suas capacidades de colonização.