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JOSÉ PEDRO VIEIRA ARRUDA JÚNIOR

ICHTHYOPLANKTON COMMUNITY IN TROPICAL ESTUARIES OF THE BRAZILIAN SEMIARID COAST DURING EXTREME EVENTS: DROUGHT AND HEAVY RAINFALL

FORTALEZA
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Dissertação apresentada à Coordenação do Programa de Pós-Graduação em Ciências Marinhas Tropicais do Instituto de Ciências do Mar, da Universidade Federal do Ceará, como requisito parcial para a obtenção do grau de Mestre em Ciências Marinhas Tropicais. Área de Concentração: Ciência, Tecnologia e Gestão Costeira e Oceânica.

Orientador: Prof. Dr. Marcelo de Oliveira Soares.

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“Get up Johnny boy, get up Johnny boy
Get up 'cause the world has left you lying on the ground
You're my pride and joy, you're my pride and joy
Get up Johnny boy because we all need you now.”

ABSTRACT

Tropical estuaries serve as growing and feeding areas for early life stages of fish that take advantage of mangroves as nursery zones. Despite the socioeconomic importance, studies of fish eggs and larvae (ichthyoplankton) in tropical estuaries are relatively scarce, especially in arid and semiarid regions. In a context of extreme precipitation events (e.g., drought and heavy rainfall) that directly influence the ichthyoplankton community, studies on the structure and dynamics of these organisms are essential to understand the role of these events, as they become severe. Therefore, this master dissertation is structured in papers with the objective of understand the ichthyoplankton communities in tropical estuaries, through systematic review (Chapter 01), and on semiarid Brazilian estuaries, during extreme events: severe drought (Chapter 02) and heavy rainfall (Chapter 03). The scientific literature highlights the relative importance of Brazil (N=19 papers) but warns about the scarcity of studies (N=1 papers) in the Brazilian semiarid coast. Water temperature (N=10 papers) and salinity (N=8 papers) are the key environmental factors that significantly affect the ichthyoplankton community in the estuaries analyzed. However, even though these ecosystems are important nursery zones for species of different origins, further research need to be done to better understand their nursery function and the effect of global environmental changes. Piranji estuary (affected by severe drought) was inverse and hypersaline most of the year (dry season) and the ichthyoplankton community presented low density, following a pattern of increasing density towards the downstream zone, near the river mouth, and low diversity, composed of few species of stress-tolerant estuarine and marine species (*Anchoa hepsetus*, *Bathygobius soporator*, *Hippocampus reidi*, *Eucinostomus* sp., *Diapterus auratus*, *Caranx latus* and *Bardiella ronchus.*, *Echeneis naucrates* and *Haemulon* sp.). In addition, the dominance of preflexion larvae may indicate difficulty of development due to the retention in the system. Pacoti estuary (affected by heavy precipitation) was positive throughout the year mainly due to the high input of freshwater, during intense rainfall and dam water release. However, the ichthyoplankton community also presented low densities and the larvae assemblage was composed of estuarine and marine species (*Atherinella brasiliensis*, *Gobionellus oceanicus*, *Sphoeroides testudineus*, *Strongylura* sp., *Hyporhamphus unifasciatus*, *Eucinostomus* sp., Sciaenidae sp1 and *Anchovia clupeoides*). Freshwater species were absent. In Pacoti, the eggs were concentrated in the downstream zone, near the river mouth, and the larvae, although distributed along the river, were concentrated in the upstream zone. Heavy precipitations events may have flushed these organisms out of the estuary and thus reduced the density of these organisms. The dominance of flexion and postflexion larvae in the upstream zone may indicate that this area is used as a feeding area for the fish larvae. Studies of ichthyoplankton in tropical estuaries can elucidate the influence of global changes and extreme precipitation events on the recruitment of commercial fishes and the nursery function of mangroves. In this research, we present novel and valuable information about these organisms that are sensitive to environmental changes and research directions that can collaborate with the ichthyoplankton monitoring and the management of tropical estuaries.

Keywords: Heavy precipitation. Fish eggs and larvae. Severe drought. Semiarid Coast of Brazil. Mangroves.

RESUMO

Os estuários tropicais servem como áreas de crescimento e alimentação para as fases iniciais de vida dos peixes que aproveitam os manguezais como berçários. Apesar da importância socioeconômica, os estudos de ovos e larvas de peixe (ictioplâncton) em estuários tropicais são relativamente escassos, especialmente em regiões áridas e semiáridas. Em um contexto de eventos climáticos extremos, os quais influenciam diretamente a comunidade do ictioplâncton, os estudos sobre a estrutura e dinâmica destes organismos são essenciais para compreender a influência destes eventos cada vez mais severos. Portanto, esta dissertação de mestrado está estruturada em artigos com o objetivo de compreender as comunidades de ictioplâncton em estuários tropicais, através de revisão sistemática (Capítulo 01), e em estuários semiáridos brasileiros, durante eventos extremos: seca severa (Capítulo 02) e chuva intensa (Capítulo 03). A literatura científica destaca a importância relativa do Brasil (N=19 artigos), mas adverte sobre a escassez de estudos (N=1 artigos) na costa semiárida brasileira. A temperatura da água (N=10 artigos) e a salinidade (N=8 artigos) são os principais fatores ambientais que afetam significativamente a comunidade de ictioplâncton nos estuários analisados. No entanto, embora estes ecossistemas sejam importantes berçários para espécies de diferentes habitats, é necessário conduzir novos estudos para melhor compreender essa função e o efeito das mudanças ambientais globais. O estuário de Piranji (afetado por seca severa) foi inverso e hipersalino na maior parte do ano (estação seca) e a comunidade de ictioplâncton apresentou baixa densidade, seguindo um padrão de densidade crescente em direção à foz do rio, e baixa diversidade, composta por poucas espécies de stress-tolerantes e marinhas (*Anchoa hepsetus*, *Bathygobius soporator*, *Hippocampus reidi*, *Eucinostomus* sp., *Diapterus auratus*, *Caranx latus* e *Bardiella ronchus*, *Echeneis naucrates* e *Haemulon* sp.). Além disso, o predomínio de larvas de pré-flexão pode indicar dificuldades de desenvolvimento devido à retenção em um sistema hipersalino. O estuário de Pacoti (afetado por fortes precipitações) foi positivo durante todo o ano, principalmente devido à elevada descarga de água doce, durante chuvas intensas. Contudo, a comunidade de ictioplâncton também apresentou densidades baixas e o conjunto de larvas era composto por espécies estuarinas e marinhas (*Atherinella brasiliensis*, *Gobionellus oceanicus*, *Sphoeroides testudineus*, *Strongylura* sp., *Hyporhamphus unifasciatus*, *Eucinostomus* sp., Sciaenidae sp1 e *Anchovia clupeoides*). As espécies de água doce foram ausentes. No Pacoti, os ovos se concentraram na região da foz do rio, e as larvas, embora distribuídas ao longo do rio, concentraram-se na zona à montante. As precipitações fortes podem ter expelido estes organismos para fora do estuário, reduzindo assim a densidade destes organismos. O predomínio de larvas de flexão e pós-flexão à montante pode indicar que esta zona é utilizada como área de alimentação para as larvas de peixe. Estudos sobre o ictioplâncton em estuários tropicais podem elucidar a influência de alterações globais e eventos de precipitação extrema no recrutamento de peixes e na função de berçário dos manguezais. Nesta investigação, apresentamos informação inédita sobre estes organismos sensíveis às mudanças ambientais e orientações de investigação que podem colaborar com o monitoramento do ictioplâncton e a gestão de estuários tropicais.

Palavras-chave: Precipitação intensa. Ovos e larvas de peixes. Seca severa. Costa Semiárida do Brasil. Manguezal.

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SUMMARY

1 GENERAL INTRODUCTION

Estuaries can function as refuge for various species and as an ecotone region essential to the life history, reproduction, and development of many marine species (KENISH, 2019). These many species of different sizes and forms take advantage of the food provided by these productive coastal environments and by the shelter structured by vegetated habitats, such as seagrass beds, salt marshes, and mangrove forests (WHITFIELD; ELLIOT, 2011). Estuaries are also important for humans, offering ecosystem goods and services, such as fisheries, coastal storm attenuation, shoreline protection, water for growing crops, and ecological tourism (YEE *et al*., 2019; BOOI; MISH; ANDERSEN, 2022).

An estuarine system is a coastal zone that has a restricted connection to the ocean and remains open at least intermittently. It can be subdivided into three regions, according to Snedden, Cable and Kjerfve (2013): **I -** a tidal river zone or fluvial zone characterized by lack of ocean salinity, but subject to tidal rise and fall of sea level; **II -** a mixing zone, reaching from the tidal river zone to the seaward location of a river mouth bar or ebb-tidal delta; and **III -** a nearshore turbid zone in the open ocean between the mixing zone and the seaward edge of the tidal plume at full ebb tide. Historically, the definition of estuaries is based on systems with constant freshwater inputs, not including the features of estuaries from hot arid and semiarid regions (ELLIOT; MCLUSKY, 2002; ELLIOT; WHITFIELD, 2011).

These unique arid and semiarid regions are fairly distributed within the surface of earth: Western and Central North America, Western and Northeast South America, Northern and Southern Africa, Australia, Arabian Peninsula, Middle East, and Western India (BECK *et al*., 2018). The climate characteristics of these regions are high solar radiation and thus high mean daytime air temperatures, overall low atmospheric humidity and rainfall, because of the predominance of high-pressure systems (descending air masses). Moreover, we can find prominent within-year rainfall seasonality, due to the north-south migration of the intertropical convergence zone, and high inter-annual variability of rainfall, often linked to variable modes of the global climate system, such as the ENSO phenomenon (KASEI, 2012; SCHOLES, 2020).

A particular characteristic of the estuaries found in the hot arid and semiarid regions is the high salinity values, most of the times even higher than the adjacent sea salinity, a condition known as hypersalinity, which results from water loss through high evaporation rates, high water residence time, and little or no input of fresh and/or sea water (POTTER *et al*., 2010; CHEVALIER *et al*.,2013; DESCROIX *et al.*, 2020). Other factor that leads to this condition in these estuarine systems is the presence of dams along the hydrographic basin, created to store and keep water for social needs, however, resulting in a reduction of the volume discharged by rivers and tributaries, which leads to an increase of salinity values (ZAFARNEJAD, 2009; KIM; KIM, 2020). Therefore, in these regions, the classical estuarine regime appears only in a few months during rainy season and there is a change in the environmental conditions in the water column of the estuarine system and nearby habitats, which directly affects the estuarine biological community (GILLANDERS *et al*., 2022; LOWE *et al.*, 2022; VAN NIEKERK *et al.*, 2022). Among the many organisms that are affected by these changes, we emphasize the plankton organisms.

Plankton organisms inhabit the water column and are passively transported by currents and winds, mainly due to their small size and low mobility (DIPPER, 2022), and because of these, the obligate wandering existence of plankton has consequences for their growth, reproduction, and evolution, as well as their distribution (BRIERLEY, 2017). Differences in temperature, nutrient concentration, food availability, competition/predation conditions along divergent trajectories have major biological impacts on plankton (BEAUGRAND, 2017; BRIERLEY, 2017). Because of its lifestyle, the plankton have a high and rapid ability to react to environmental changes, which makes them key bioindicators, especially in estuaries, where abiotic factors fluctuate drastically (FERDOUS; MUKTADIR, 2009; MACMANUS; WOODSON, 2012).

Estuarine plankton are taxonomically diverse, composed of protozoa, plants, animals, fungi, bacteria, and viruses, with each of these components playing an important role in the ecosystem functioning (BRIERLEY, 2017). The zooplankton community is composed of animals that spend their entire life cycle in the plankton (holoplankton) and eggs and larvae of invertebrates and fish, which, in turn, spend only part of their life cycle in the plankton (meroplankton) (RICHARDSON, 2008; GARCIA *et al.*, 2021). Fish eggs and larvae, commonly known as ichthyoplankton, are an economically important component of the meroplankton, since the abundance of adults in fish stocks is directly related to conditions in the plankton that favor the complete development of the larvae (HOUDE, 2019; HOUDE *et al*., 2022). Moreover, like other zooplankton organisms, planktonic fish larvae play a keystone role at the base of the trophic webs as a link between lower and higher trophic levels, since they act as both predator and prey (MONTAGNES; DOWER; FIGUEIREDO, 2010; STEINBERG; LANDRY, 2017).

Ichthyoplankton surveys aim to elucidate the causes of recruitment variability on fish stocks, the effects of habitat change through human pressure and invasive species, and to better identify, monitor and manage the spawning and nursery zones (MILLER; KENDAL JR, 2009; ZACARDI *et al.*, 2020). In estuaries, the ichthyoplankton community is widely studied to understand how the structure of the assemblage varies in relation to environmental fluctuations, as well as the behavior of these organisms in the face of these drastic variations (LIMA, FERREIRA, BARLETTA, 2019; GUERRERO *et al.*, 2021).

These environmental variables are also affected by the expansion of urban developments, disposal of untreated sewage and the degradation of essential habitats that intensify the loss of the nursery function of estuaries (TOFT *et al.*, 2018; TAYLOR; SUTHERS, 2021). Indeed, for ichthyoplankton, these adverse conditions alter the trophic structure of the estuarine system and lead to changes in the survival, diversity and distribution of fish larvae (CHU *et al*., 2021; QUAH *et al.*, 2021; RODRIGUES *et al.*, 2022).

Early life stages of fishes are also very sensitive to tidal variation and freshwater inflow, since these factors affect the physical, chemical, and biological parameters of the water column (STRYDOM, 2014; RAMOS; PARIS; ANGÉLICO, 2017; ZHANG *et al.*, 2022). Among these environmental variables, salinity shows up as a fundamental parameter in the semiarid region by structuring the ichthyoplankton assemblages in estuaries (WHITFIELD, 2015; SANTOS; RAMOS; BONECKER, 2017). Changes in salinity values in estuaries worldwide are expected due ongoing climate change and their extreme weather events, which brings organisms, such as fish eggs and larvae, to the edge of the environmental thresholds (NICHOLSON *et al.*, 2008; HALLET *et al*., 2017; ELLIOT *et al.*, 2019). Therefore, the hypersaline condition in estuaries may be more persistent and accentuated worldwide, especially in estuaries located in arid and semiarid regions (GILLANDERS *et al*., 2011; WHITFIELD, 2021).

The influence of this condition in the ichthyoplankton community is well studied in temperate estuaries of South Africa (STRYDOM, WHITFIELD, WOOLDRIDGE, 2002; WHITFIELD, 2005; CYRUS *et al.*, 2011; STRYDOM, 2014), which gives us important information on the structure and survival of fish eggs and larvae. However, studies are still scarce in tropical regions, where the condition of shallow estuaries and low river flow in arid and semiarid regions results in a naturally-occurring hypersaline systems.

Mangroves of tropical estuaries are key environments for the successful development of early life stages of fish, especially due to the presence of mangrove roots that act as nursery zones by providing shelter against predation and food in the form of organic matter exported into the water column (VORSATZ; PATTRICK; PORRI, 2021; TARIMO *et al.*, 2022). However, the number of published studies in tropical estuaries is still relatively lower compared to studies in temperate systems (ZHANG *et al.*, 2022), which hinders the knowledge about the structure of ichthyoplankton in tropical estuaries, as well as the factors that influence this community.

In the Southwest Atlantic, tropical estuaries with seasonal hypersalinity events (sal. > 37) are found on the Semiarid Coast of Brazil (SCB), which comprises a geomorphological sector of the states of Maranhão, Piauí, Ceará and Rio Grande do Norte (Northeast Brazil) (SCHETTINI *et al.*, 2017; VALENTIM; MENEZES; TEIXEIRA, 2018; SOARES *et al.*, 2021). Research on the influence of hypersalinity on the ichthyoplankton community in these tropical estuaries is of great importance, as it can reduce the scarcity of studies in tropical systems, as well as collaborate with the prediction of future scenarios, since factors that result in this condition have intensified worldwide (WETZ; YOSKOVICH, 2013; MING *et al.*, 2021; LEAL FILHO *et al.*, 2022). Thus, based on what was discussed above, this master's thesis aims to develop:

I - A systematic review about the structure of ichthyoplankton assemblages in tropical estuarine systems and aspects relate to scientific production, sampling methodology, and the use of mangroves for early life stages of fish relate to environmental variables.

II - An analysis of an intra-annual distribution and diversity of fish eggs and larvae in the tropical estuary of Piranji river (Semiarid Coast of Brazil), during a year (2015) of severe drought and extreme salinity values (hypersalinity).

III - An analysis of an intra-annual composition and spatial distribution of fish eggs and larvae in the tropical estuary of the Pacoti River (Semiarid Coast of Brazil) during heavy precipitation events throughout the 2022 year.

Based on these aims, we provide novel information about the ecology of ichthyoplankton in tropical estuarine systems, highlighting ecological and methodological aspects, and trying to elucidate the influence of contrasting extreme events (severe drought vs. heavy precipitation) on the structure of fish eggs and larvae, using tropical mangroves from the Semiarid Coast of Brazil as a model.

MSC. DISSERTATION STRUCTURE

Chapter 01 - Structure of fish eggs and larvae in tropical estuaries: key findings, gaps, and future directions.

Objectives

Compile and analyze scientific literature about the ichthyoplankton assemblages in tropical estuaries and the influence of environmental variables.

Hypothesis

The literature on this topic is relatively low compared to studies in temperate regions. Furthermore, although salinity and temperature are the main drivers of the ichthyoplankton community in tropical estuaries, the lack of data standardization prevents comparison.

Chapter 02 - Analysis of a hypersaline drought-prone estuary reveals low density and diversity of fish eggs and larvae.

(Submitted in August/2022 in the journal **Marine Pollution Bulletin** and published in January/2023)

Objectives

Analyzed the intra-annual distribution and density of fish eggs and larvae in a tropical estuary (Semiarid Coast of Brazil), during a year of severe drought.

Hypothesis

The density of fish eggs and larvae is drastically reduced, and the larvae assemblage is only composed of stress-tolerant estuarine and marine species.

Chapter 03 – Composition and estuarine distribution patterns of early life stages of tropical fish under seasonal precipitation

Objective

Analyze the structure and distribution of fish eggs and larvae in a tropical estuary of the Semiarid Coast of Brazil, during different precipitation regimes (rainy, transition and dry) in a year of heavy precipitation events.

Hypothesis

The density of fish eggs and larvae is lower during the rainy season in relation to the other seasons, due to the heavy precipitation that carry organisms out of the estuary and ecological disturbance due episodic and heavy precipitation. Although precipitation reduces the salinity of the estuary, the larvae assemblage is composed only by a few resident euryhaline and marine species.

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CHAPTER 01 - Fish eggs and larvae in tropical estuaries: key findings, gaps, and future directions

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HIGHLIGHTS

- The annual publication of fish eggs and larvae ecology in tropical estuaries is low.
- Few surveys in tropical estuaries from arid and semiarid regions.
- Water temperature and salinity variation affect the nursery function of mangroves.
- A comparison between the ecosystems can only be made when the sampling volume is standardized.
- The use of more than one mesh size to sample should be tested.

ABSTRACT

Estuaries are important environments that act as nurseries for fish eggs and larvae. Despite their importance, most studies have been done in temperate estuaries. In this context, understanding the current state of knowledge on fish eggs and larvae in tropical estuaries (e.g., mangroves) is fundamental for the development of conservation strategies. In this sense, the scientific production, ecological and methodological sampling of these tropical assemblages were accessed. Overall, we analyze 31 papers that surveyed tropical estuaries from the following biogeographical provinces: Tropical Atlantic (n=23), Tropical Eastern Pacific (n=1), Western Indo-Pacific (n=3), Central Indo-Pacific (n=4). The large number of Tropical Atlantic studies is due to the studies in South America, mainly in Brazil, where the Semiarid Coast of Brazil is neglected (n=1 paper). Low number of publications per year (1-5) were published between 1988-2022, with a peak in the 2017 year. Most of the studies were published in Estuarine, Coastal and Shelf Science (15% of the publications). Regarding the types of net and mesh sizes, Bongo net (32% of the total publications) is the main net type used for sampling, as well as the mesh size of 500 μm (48%). However, the methodological approach that more than one net mesh size better explains the diversity of mangroves by fish eggs and larvae must be tested. Through the listing of the most abundant species, we highlight the use of mangroves as nursery areas for freshwater, estuarine, and marine species. Temperature and salinity are the main factors mentioned as structuring these tropical estuarine assemblages. Therefore, changes in these factors due to climate change should be monitored, as the function of these mangrove estuaries as nursery zones may be compromised. Although these studies present important information, the non-standardization of the sampling volume for calculating the fish eggs and larvae abundance prevents spatial comparison worldwide. This review contributes to the development of research, especially in a context of ongoing climate change and local human pressures as increasing threats to tropical estuaries.

Keywords: Estuaries. Fish eggs. Fish larvae. Mangroves. Nursery zones. Salinity.

1 INTRODUCTION

Studies on early life stages of fishes had great attention with the development of the Hjort's (1914) hypothesis, which proposed that the recruitment variability of fishes is related to prey consumption (Critical Period hypothesis) and advection of fish eggs and larvae (Aberrant Drift hypothesis) (HOUDE, 2008). Studies on this topic have been the emphasis in trying to describe the variability in recruitment rates, since their early development directly influences the adult fish stocks. As a result, the economic and conservation appeal has spurred research groups worldwide to seek to describe the patterns of distribution and diversity of fish eggs and larvae at different scales of time and space. Great attention has been paid, especially in a context of global environmental changes caused by extreme weather events and human impacts such as microplastics and oil spill (CRUZ, PAIVA. GOMES, 2016; RODRIGUES *et al.*, 2019; VAGNER, ZAMBONINO-IFANTE, MAZURAI, 2019; SOUZA *et al.*, 2022).

Ichthyoplankton assemblages are studied worldwide, in different perspectives, from freshwater habitats to oceanic islands. Among the various ecosystems studied, coastal wetlands, such as estuarine systems, are widely recognized as being important nursery zones for early life stages of fish and other species (FRANTINE-SILVA *et al.*, 2015; POTTER *et al.*, 2015; LIMA, FERREIRA, BARLETTA, 2019). In these systems, freshwater, brackish and marine species of fish larvae take advantage of habitat heterogeneity and food supply to develop and grow (OOI; CHONG, 2011; WHITFIELD, 2020). Furthermore, diadromous fish species migrate between the freshwater and marine environments using the estuarine system as an ecological corridor (HUANG, 2019; CRESCI, 2020).

Tropical estuaries are bordered along its length by mangrove forests, where local fisheries benefit from the mangrove-associated species that uses this environment as feeding and breeding zones (WHITFIELD, 2017; HABIB *et al.*, 2020; FIERRO-ARCOS *et al.*, 2021; SEARY *et al.*, 2021). Mangroves also play a key role as nursery zones for target economic species, due to the input of nutrients that improves the estuarine food webs, and the mangrove creeks and roots that provides a less turbulent and protected habitat against predation (ABRANTES *et al.*, 2015; TARIMO *et al.*, 2022).

Nursery areas have suitable environmental conditions and the possibility of connectivity between habitats that allow maximum recruitment for fishes (BECK *et al.*, 2001; SHEAVES *et al.*, 2015; GUERRERO *et al.*, 2021; SWADLING *et al.*, 2022). However, even though a typical feature of the tropical zone is the low intra-annual temperature variability, tropical estuaries have particularities related to their hydrological (e.g, tidal regimes, and input of freshwater), climatic and geomorphological characteristics, which directly affects the abundance and distribution of fish eggs and larvae (BLABER, 2002; SONG, ZHANG. XIAXIANG, 2019; HOUDE *et al.*, 2022).

Potential effects of the ongoing climate change on early life stages of fish have been called to the attention of researchers, especially to reveal the environmental changes that these key organisms may undergo (GUILANDERS *et al.* 2011; BLABER, 2013). The increased sea surface temperature and the frequency of extreme precipitation events has been constantly reported by the IPCC (Intergovernmental Panel on Climate Change), and trends of water scarcity, erratic or exacerbated rainfalls and rising marine heatwaves in tropical regions will probably affect the ichthyoplankton assemblage (MOHAMMED, SCHOLZ, 2019; HALIK, PUTRA, WIYONO, 2020; SCHOLES, 2020; SINGH; CHUDASAMA, 2021).

The literature about the estuarine ichthyoplankton is more concentrated in the temperate region than in tropical estuaries (STRYDOM, 2015; ZHANG *et al.*, 2022). This knowledge gap limits the information on fish and eggs assemblages and the climate change impacts. Moreover, restricts the development of mitigation and adaptation plans in tropical countries. Therefore, we compile and analyze the scientific production on the structure of fish eggs and larvae in tropical estuaries, analyzing the adopted sampling methodologies and knowledge gaps on community ecology and environmental variables.

With the main findings of this paper, we hope that we can (I) elucidate the panorama of scientific production on the subject for the tropical estuarine region; (II) highlight the influence of estuarine environmental variables on the structure of fish eggs and larvae and the importance of mangroves as nursery zones; (III) discuss aspects of sampling and quantification of fish eggs and larvae and, (IV) point future directions on the tropical estuarine ichthyoplankton research.

2 MATERIAL AND METHODS

Searches for peer-reviewed articles were conducted using four most common bibliographic databases; Scopus, Web of science, ScienceDirect, and Google Scholar. Search terms used with Boolean operators and wildcard '*' to identify articles available. The wildcard is not supported only in ScienceDirect. The term (Ichthyoplankton) OR (Fish eggs) AND (Fish larvae) AND (Estuary OR Estuaries OR Estuarine) AND (Tropical) was used to select publications with these terms in the title, abstract and keywords, simultaneously. The list with the publications used in this review is presented as supplementary material I. Only data published between 1988 – 2022 in Portuguese, English and Spanish were considered. We did not include gray literature [e.g, academic works (e.g., dissertation, thesis), conferences, books, and book chapters] in this review.

We used the standardized Rayyan methodology (Intelligent Systematic Reviews) (JOHNSON; PHILLIPS, 2018) and PRISMA (Preferred Reporting Items for Systematic Reviews and Meta Analysis) (O'DEA *et al.*, 2021) to conduct the literature compilation and selection. Titles and abstracts of each study were independently filtered according to the eligibility criteria by three of the authors. We included publications where studies were conducted in tropical estuaries and analyze the ecological aspects of the ichthyoplankton assemblage (e.g., density and diversity) and/or estuarine environmental variables. We did not include publications that were restricted to only one species or that did not present data regarding environmental variables on tropical estuaries. The full article was retrieved when decisions were unable to be made from abstract and title alone. We selected publications after exclusion of duplicates and inclusion of publications based on the inclusion criteria. The information extracted from the papers is described in Table 1.

Table 1 – Information extracted from published papers included in this review on ichthyoplankton in tropical estuaries between the years 1988 - 2022. The results comprise scientometrics information, sampling apparatus, diversity and quantification of fish eggs and larvae, and environmental variables.

Group of information	Extracted information
Scientometrics analyses	Country, year of publication, and scientific journal where the study was published.
Methodological aspects	Type of nets and mesh sizes of nets to sampling fish eggs and larvae.
Diversity	Most abundant fish taxa and ecological guilds.
Abundance/Density	Quantification of fish eggs and larvae.
Environmental conditions	Tested environmental variables that affect the estuarine ichthyoplankton assemblages.

The scientific production on the topic of this review in each country was summarized in the biogeographic regions proposed by Spalding *et al.* (2007), which was based on coastal and shelf waters, combining benthic and shelf pelagic (neritic) biotas. A time series of years in which each study was published was done to highlight the number of publications in each year and when there were more studies published; the relative percentage (%) of the journals where these studies were published were represented by pie charts, as well as, to represent the type of net and mesh sizes used for sampling ichthyoplankton; a bar graph was used to represent the families of the most abundant species and the number of mentions with which some environmental variable was mentioned as positively or negatively influencing the ichthyoplankton assemblages. The mentions mean the number of papers where a variable or taxon was cited. All these analyses were done using the Excel program's graphing tool.

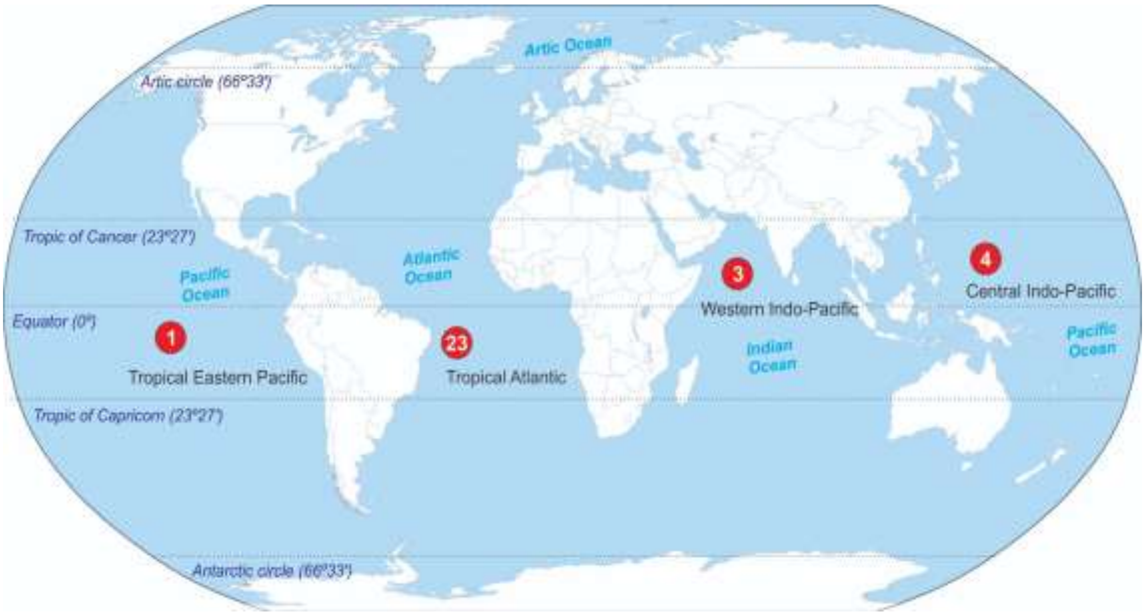
We also built two important tables: the first one that highlights the most abundant species in each study, as well as the ecological guilds they belong to (freshwater, estuarine, and marine), and the second one that presents data from volume units that are used to express the density of the organisms.

3 RESULTS

3.1 Scientometrics data

Through the search method used, we found 435 publications, 340 from ScienceDirect, 28 from Web of Science, 20 from SCOPUS and 44 from Google Scholar between 1988 and 2022, resulting in 31 papers included in this review. These studies belong to four biogeographic provinces: *Tropical Atlantic* (TA), with 23 papers from Brazil (N=19), Colombia (N=1), French Guiana (N=1), Mexico (N=1) and Senegal (N=1); *Tropical Eastern Pacific* – TE, with only one paper from Colombia; *Western Indo-Pacific* – WIP, with 3 papers from India (N=1), Kenya (N=1) and Iran (N=1); and *Central Indo-Pacific* – CIP, with 4 papers from Malaysian (N=2) and Indonesian estuaries (N=2). (Figure 1).

Figure 1 – Biogeographic provinces of the tropical zone where are located the studied and analyzed tropical estuaries of the papers included in this review (1988 to 2022), indexed in Scopus, Web of Science, ScienceDirect and Google Scholar databases, highlighting the number of publications for each of the provinces proposed by Spalding *et al.* (2007).

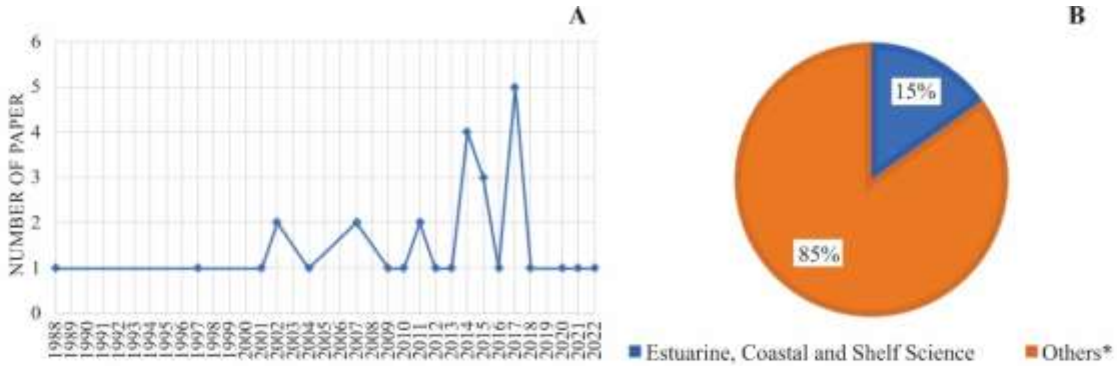


Estuaries from the Tropical Atlantic province covers the systems from Mexico to Brazil, in addition to the hypersaline mangroves of Senegal (West African coastline). Among the countries of this biogeographic province, Brazil contributed the most to this review, with 19 papers. The Brazilian coastline can be divided into Marine Ecoregions (SPALDING *et al.* 2007) such as: *Amazon ecoregion*, in the Northern Brazil Shelf, with 8 studies; *Northeast and Eastern Brazil ecoregions*, with 11 studies, in the Tropical Southwestern Atlantic. Furthermore, it is important to note that only one study was found in a region situated in the *Northeastern ecoregion*, known as the semiarid coast of Brazil. This study was conducted in the Tubarão e Casqueira estuaries, in the brazilian state of Rio Grande do Norte.

The analysis of the temporal distribution (1988 - 2022) of these papers reveal that these studies about fish eggs and larvae in mangroves (tropical estuarine systems) have a low annual production. Although we can see an increase in the number of papers per year and a peak of publication in the year of 2017, this number does not exceed 5 articles per year and has a reduction of the annual publication onward, following the period of the COVID-19 pandemic that affected the world's scientific production (Figure 3a).

Regarding the journals where these studies were published, 26 journals contributed for this review with Estuarine, Coastal and Shelf Science accounting for 5 papers (15% of the total papers), which was the journal that contributed the most to the studies. The other journals totaled 26, or 85% of the other journals. (Figure 3b).

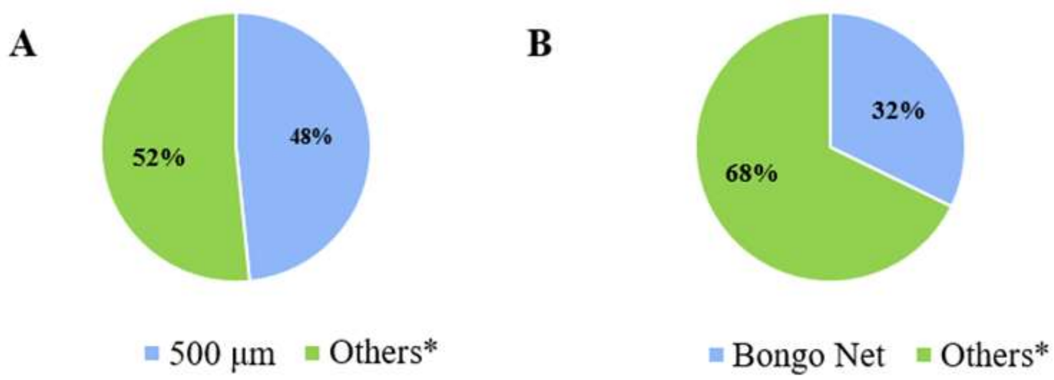
Figure 2 – Temporal analysis (1988-2022) and journals (scientific production) where the 31 studies included in this review were published. **A** - Analysis of the temporal distribution, between the years 1988 and 2022, in which the analyzed studies were published. **B** - Journals where the studies were published, with emphasis on Estuarine, Coastal and Shelf Science that recorded most of the papers. Others* are the other journals where the studies were published.



3.2 Type of nets and mesh sizes of nets

Different types of nets (bongo, conical, conical-cylindrical, square frame net with a conical codend, drag and trap, ring trawl and neuston - combined and Unspecified) and mesh size (µm) of nets (158, 200, 300, 303, 330, 333,450, 500 and combined use of 200 and 300, 180 and 363, 330 and 500) were used to sample mangrove-estuarine ichthyoplankton (Figure 4A and 4B). The 500 µm mesh size alone was the most selected to sample ichthyoplankton (48% of the total publications). The other sizes and combinations of sizes accounted for 26%. Among the types of nets utilized in these studies, Bongo (32%) were the main types utilized. The other 5 types of nets accounted for 68%.

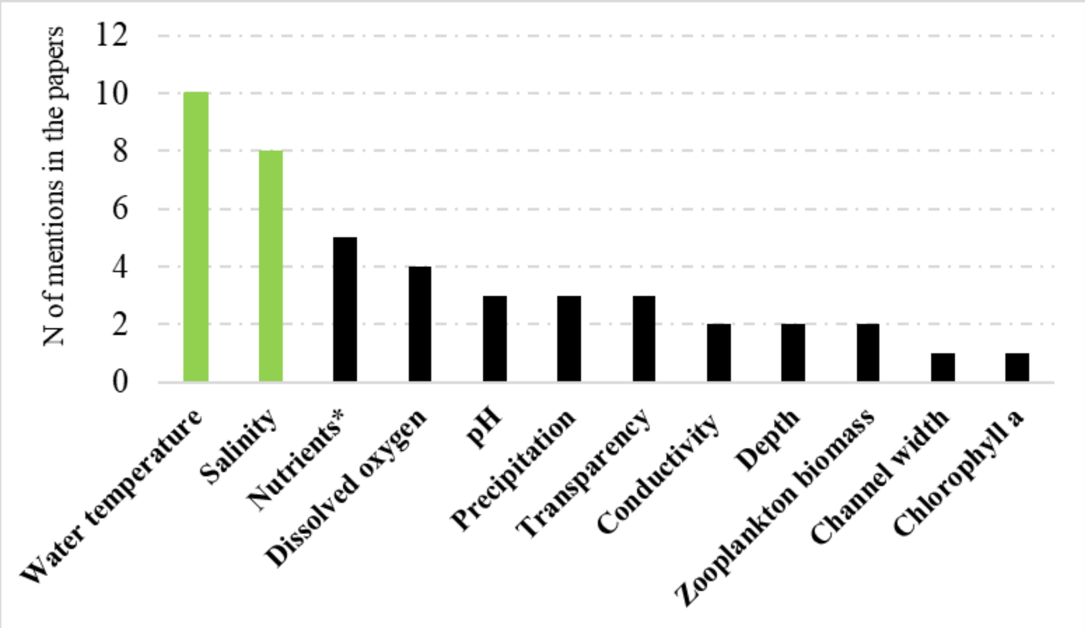
Figure 03 – Types of net and mesh size (sampling apparatus) used for sampling ichthyoplankton in the tropical estuaries analyzed in this review. The percentage (%) indicates the ratio of net type or mesh size among the total number of net types and mesh size (s) used in the studies. **A** - Relative percentage of mesh sizes of nets utilized in these studies, with high percentage of 500 μm mesh size **B** - Relative percentage of each type of net utilized in the studies, with high percentage of bongo among the net types. Asterisk (*) means the less abundant types and sizes.



3.3 Environmental variables and estuarine ichthyoplankton

Among the 31 papers analyzed in this review, a total of 19 performed some analysis aimed at determining which estuarine variables have some influence (positive or negative) on some ecological aspect of the fish eggs and/or larvae. These studies cited the influence of 12 variables as determining the structure of this assembly in the estuarine environment. Figure 5 shows the number of papers where these variables were mentioned with emphasis on Water temperature (N = 10) and salinity (N = 8).

Figure 4 – Environmental variables influencing the structure of fish egg and larval assemblages in tropical estuaries presented and tested in the studies (1988 - 2022). Emphasis should be placed on water temperature and salinity. Asterisk (*) in nutrients means we include organic and inorganic nutrients as one.



3.4 Fish eggs and larvae quantification and families of the most abundant species

In the analyzed studies, fish eggs and larvae were quantified to show spatial-temporal variations and relationships with the estuarine environmental variables. Most of the studies used density (Number of organisms in a given volume – N/V) to quantify the assemblage in the estuaries analyzed (2 studies only presented the number of eggs and larvae). However, the density values are not standardized across studies (e.g., 1 study used ‘m^2’ instead of ‘m^3’) (Table 01). This lack of uniformity among the data makes it difficult to make spatial quantitative comparisons among tropical estuaries and prevents the scientific community from understanding the particularities of each system, as well as the influence of the environmental drivers.

Table 02 – Density/Abundance units used to analyze fish eggs and larvae density in tropical estuaries. The studies were divided according to the biogeographic provinces proposed by Spalding *et al.* (2007). Below is highlighted the units in which the densities of fish eggs and larvae were standardized. Some studies used standard error or standard deviation (±SD or SE) to express the data, while others only expressed the number of organisms (N). Note that one study used the unit m^2(underlined).

Biogeographic provinces	**Unit**	**Number of papers**
Tropical Atlantic	Number of ind./50m³	1
	Number of ind./100m³	16
	Number of ind./100m³±SD	4
	Number of fish eggs and larvae (no density)	2
Tropical Eastern Pacific	Number of ind./1000m³	1
Western Indo-Pacific	<u>Number of ind./10m²</u>	1
	Number of ind./100m³±SD	1
	Number of ind./100m³	1
Central Indo-Pacific	Number of ind./100m³±SE	1
	Number of ind./100m³	3

The dominant taxa belong to 11 orders and 27 families (Table 02). The identified species have varied origins, coming from freshwater, estuarine and marine environments (reef, coastal, shelf). These results indicate that mangroves are target areas for the development and survival of these organisms until adulthood. Among the families described in the studies, species from the families Engraulidae (most studies), Gobiidae, Clupeidae and Sciaenidae were frequently mentioned as the most abundant taxa, as shown in Figure 06.

Table 3 – Species of early stages of fish larvae most abundant in the mangroves (tropical estuaries) studied in this review in the columns are presented the common name of these species, the ecological guilds to which they belong (according to the preferred habitat of adults) and the studies where these species were most abundant. Note that "only family*" means that the author has not identified the species beyond the family, which makes it difficult to find more precise information. Some genus-level taxa were classified according to ecological guilds. E = estuarine, F = freshwater, M = marine.

Species	Common name	Ecological guilds	Studies where the taxa were dominant in the estuarine system
ATHERINIFORMES			
Atherinopsidae			
Atherinella brasiliensis	Brazilian silverside	E	Badú, Lima e Pessanha (2021)
Atherinella blackburni	Beach silverside	E	Correa-Herrera et al. (2017)
BELONIFORMES			
Hyporhamphus sp.	-	E, M	Correa-Herrera et al. (2017)
Hyporhamphus picarti	African Halfbeak	E, M	Sloterdijk et al. (2017)
BLENNIIFORMES			
Blenniidae			
Only family*	-	-	Arshad et al. (2012); Ara et al. (2020)
Hypleurochilus langi	Combtooth blenny	E, M	Sloterdijk et al. (2017)
Hypsoblennius invemar	Tessellated blenny	M	Marcolini et al. (2010)
Labrisomidae			
Only family*	-	-	Lopez and Arias (1986)

CHARACIFORMES			
Characidae			
Astyanax sp.	-	F	Correa-Herrera et al. (2017)
CLUPEIFORMES			
Clupeidae			
Only family*	-	-	Blaber et al. (1997); Chermahini et al. (2011); Costa et al. (2011); Arshad et al. (2012); Zacardi, Sobrinho, and Silva (2014); Sloterdijk et al. (2017)
Harengula clupeola	False Herring	E, M	Lima, Barletta and Costa (2015)
Harengula jaguana	Scale Herring	E, M	Marcolini et al. (2010)
Lile piquitinga	Atlantic Piquitinga	E, M	Badú, Lima e Pessanha (2021)
Rhinosardinia amazonica	Amazon Spinejaw Sprat	F, E	Zacardi. Bittencourt and Nakayama (2016)
Rhinosardinia bahiensis	Bahia Sprat	F, E	Lima et al. (2015); Lima, Barletta and Costa (2015)
Spratellomorpha bianalis	Two-finned round Herring	E	Little, Reay and Grove (1988)
Engraulidae			
Only family*	-	-	Blaber et al. (1997); Ekau et al. (2001); Joyeux, Pereira and Almeida (2004); Contente et al. (2007); Bonecker, Castro and Bonecker (2009); Costa et al. (2011); Ooi and Chong (2011); Sarpedonti, Anunciação and Bordallo (2013);

			Zacardi, Sobrinho and Silva (2014); Zacardi and Bittencourt (2016); Ara et al. (2020)
Anchoa sp.	-	E, M	Ekau et al. (2001); Marcolini et al. (2010); Correa-Herrera et al. (2017)
Anchoa hepsetus	Broad Striped Anchovy	E, M	Arévalo-Frias and Mendonça (2011)
Anchoa mitchilli	Bay Anchovy	E, M	Arévalo-Frias and Mendonça (2011)
Anchovia clupeoides	Zabaleta Anchovy	E, M	Barletta-Bergan, Barletta, and Saint-Paul (2002a); Barletta-Bergan, Barletta and Saint-Paul (2002b); Lima et al. (2015); Correa-Herrera et al. (2017)
Anchoviella spp.	-	E, M	Santos, Ramos and Bonecker (2017)
Anchoviella lepidentostole	Broadband Anchovy	E	Bonecker et al. (2007); Rousseau, Blanchard and Gardel (2017); Santos, Ramos and Bonecker (2017)
Anchoviella guianensis	Guyana Anchovy	F, E	Rousseau, Blanchard and Gardel (2017)
Cetengraulis edentulus	Atlantic Anchoveta	E, M	Lima et al. (2015); Correa-Herrera et al. (2017)
Cetengraulis mysticetus	Pacific Anchoveta	E, M	Medina-Contreras et al. (2014)
Thryssa sp.	-	E	Balakrishnan et al. (2015)
Pristigasteridae			
Only family*	-	-	Zacardi, Sobrinho and Silva (2014)
Pellona flavipinnis	Yellowfin river Pellona	F	Zacardi, Bittencourt and Nakayama (2016)
GOBIIFORMES			

Eleotridae			
Only family*	-	-	Zacardi, Sobrinho and Silva (2014)
Dormitator maculatus	Fat sleeper	F, E	Correa-Herrera et al. (2017)
Eleotris sp.	-	-	Rousseau, Blanchard and Gardel (2017)
Guavina guavina	Guavina	F, E	Barletta-Bergan, Barletta and Saint-Paul (2002b)
Gobiidae			
Only family*	-	-	Little, Reay and Grove (1988); Blaber et al. (1997); Ekau et al. (2001); Bonecker, Castro and Bonecker (2009); Costa et al. (2011); Chermahini et al. (2011); Ooi and Chong (2011); Arshad et al. (2012); Sarpedonti, Anunciação and Bordallo (2013); Sloterdijk et al. (2017); Ara et al. (2020)
Bathygobius curacao	Nochtonge Goby	E, M	Correa-Herrera et al. (2017)
Ctenogobius boleosoma	Darter Goby	E	Marcolini et al. (2010); Santos and Severi (2018)
Ctenogobius stigmaticus	Marked Goby	E, M	Rousseau, Blanchard and Gardel (2017); Santos and Severi (2018)
Gobiomorus dormitor	Bigmouth sleeper	F	Arévalo-Frias and Mendonça (2011)
Gobiosoma sp.	-	-	Zacardi and Bittencourt (2016); Correa-Herrera et al. (2017)
Gobionellus oceanicus	Highfin Goby	E	Santos and Severi (2018)
Microgobius meeki	Meek’s goby	E, M	Joyeux, Pereira and Almeida (2004); Santos and Severi (2018)

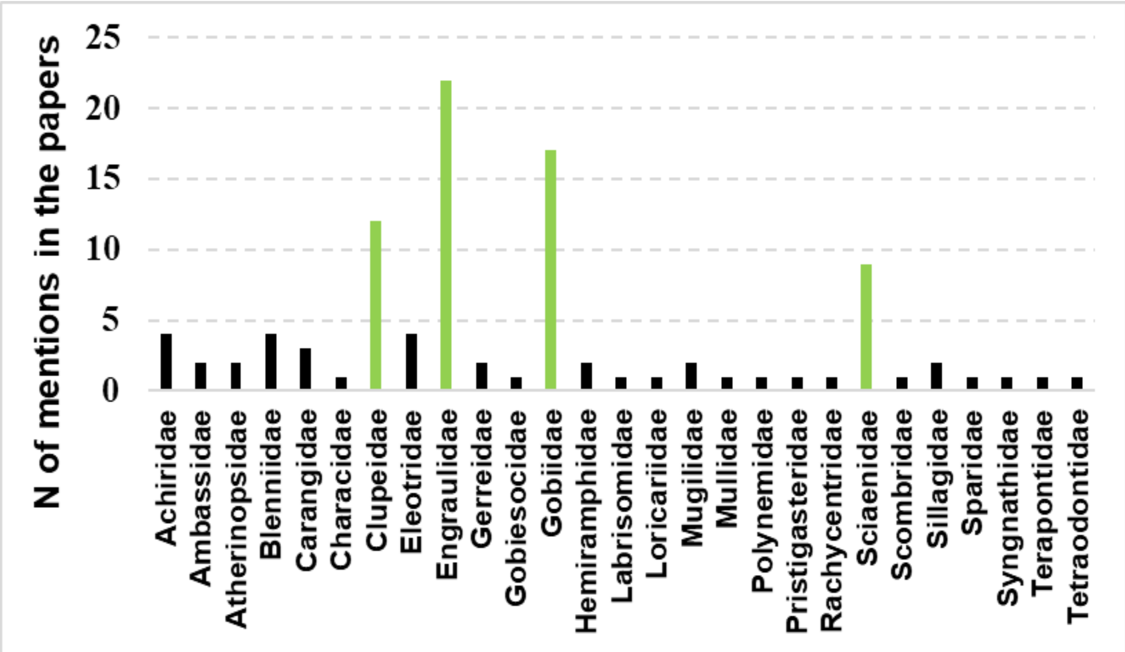
Rachycentridae			
Only family*	-	-	Ara et al. (2020)
MUGILIFORMES			
Only family*	-	-	Sloterdijk et al. (2017)
Mugil cephalus	Flathead grey Mullet	E, M	Balakrishnan et al. (2015)
PERCIFORMES			
Ambassidae			
Only family*	-	-	Ara et al. (2020)
Ambassis commersonii	Commerson's glassy	F, E, M	Balakrishnan et al. (2015)
Carangidae			
Caranx latus	Horse-eye Jack	E, M	Badú, Lima e Pessanha (2021)
Oligoplites saurus	Leatherjacket	E, M	Zacardi, Bittencourt and Nakayama (2016); Badú, Lima e Pessanha (2021)
Seriola sp.	Amberjacks	-	Medina-Contreras et al. (2014)
Gobiesocidae			
Only family*	-	-	Lopez and Arias (1986)
Sciaenidae			
Only family*	-	-	Blaber et al. (1997); Bonecker, Castro and

			Bonecker (2009); Costa et al. (2011); Sarpedonti, Anunciação and Bordallo (2013)
Bairdiella ronchus	Ground croaker	E, M	Santos and Severi (2018)
Menticirrhus americanus	Southern Kingcroaker	M	Correa-Herrera et al. (2017)
Micropogonias furnieri	Whitemouth croaker	E, M	Santos, Ramos and Bonecker (2017)
Plagioscion squamosissimus	South American Silver croaker	F, E	Zacardi, Bittencourt and Nakayama (2016)
Stellifer microps	Small eye stardrum	E, M	Barletta-Bergan, Barletta and Saint-Paul (2002a)
Stellifer rastrifer	Rake stardrum	E, M	Santos and Severi (2018)
Stellifer stellifer	Little croaker	E, M	Santos and Severi (2018)
Gerreidae			
Only family*	-	-	Joyeux, Pereira and Almeida (2004); Sloterdijk et al. (2017)
Loricariidae			
Pterygoplichthys spp.	-	F	Arévalo-Frias and Mendonça (2011)
Mullidae			
Only family*	-	-	Ara et al. (2020)
Polynemidae			
Only family*	-	-	Blaber et al. (1997)
Teraponidae			

Only family*	-	-	Arshad et al. (2012)
Terapon jarbua	Jarbua Terapon	F, E, M	Balakrishnan et al. (2015)
Scombridae			
Scomberomorus sp.	-	-	Balakrishnan et al. (2015)
Sillaginidae			
Only family*	-	-	Chermahini et al. (2011); Arshad et al. (2012)
Sparidae			
Diplodus bellottii	Senegal seabream	M	Sloterdijk et al. (2017)
PLEURONECTIFORMES			
Achiridae			
Achirus sp.	-	-	Santos and Severi (2018)
Achirus lineatus	Lined Sole	E, M	Zacardi, Bittencourt and Nakayama (2016)
Trinectes sp.	-	-	Marcolini et al. (2010); Santos and Severi (2018)
Trinectes maculatus	Hogchoker	E, M	Lima, Barletta and Costa (2015)
TETRAODONTIFORMES			
Tetraodontidae			
Only family*	-	-	Blaber et al. (1997)
SYNGNATHIFORMES			
Syngnathidae			

Only family*	-	-	Blaber et al. (1997)

Figure 5 – Families of the dominant fish eggs and larvae determined by the studies included in this review. In total, 27 families are described as utilizing the mangroves of the estuaries reviewed during early stages of development. Engraulidae, Gobiidae, Clupeidae Sciaenidae stand out among these families with a higher number of dominant species.



4 DISCUSSION

The tropical estuaries analyzed in this review were distributed along four biogeographic provinces, but most of the studies were conducted in the Tropical Atlantic province, where Brazil contributed to the most with papers. The ichthyoplankton assemblages of these systems are mainly influenced by water temperature and salinity that drives the composition and the abundance of these organisms. Species of the families Engraulidae, Gobiidae, Clupeidae and Sciaenidae are the main groups that use these systems as nursery zones, but this use is threat by global changes and human impacts. Based on the sampling methods and results of the papers analyzed, we reinforce the need to standardize data and to test hypotheses that can improve the understanding of ichthyoplankton in tropical estuaries.

4.1 Global distribution and features of the tropical estuaries analyzed

Overall, the low number of papers found in this review could be associated to the limitations found for researchers in tropical countries where this studies were conducted. Much of the studies in tropical countries related to fish science were difficult in the past due to some issues, like the lack of adequate research infrastructure and funding (BLABER; BARLETTA, 2016). Although this problem is being solved, especially in countries like Brazil, for the ichthyoplankton, problems of investment in science are still common in developing countries located in the tropical region (ZHANG *et al.,* 2022). Although it has relatively increased in the last 10 years compared to previous years, the annual scientific production is still low.

The global distribution of these studies highlights the contribution of Tropical Atlantic estuaries, in large river-dominated estuaries from Mexico, French Guiana, Colombia and Northern and Southwest Brazil. These kind of systems are also found in Pacific Colombian (TEP), Indonesian (WIP), Malaysian (WIP) and Indian (CIP). They have strong seasonal freshwater discharge that influences the psycho-chemical variables of the water column and, consequently, the diversity and abundance of ichthyoplankton (BONECKER *et al.,* 2007; ARÉVALO-FRIAS AND MENDONZA – CARRANZA, 2011; BALAKRISHNAN *et al.*, 2015; CORREA – HERRERA *et al.,* 2017). During the rainy season, freshwater discharge controls fish larvae abundance and diversity, since the currents flushes fish larvae out of the system to the sea and prevents marine species from entering the estuarine system (BARLETTA – BERGAN, BARLETTA AND SAINT – PAUL, 2002a, ROUSSEAU *et al.*, 2017).

Conversely, low-inflow and freshwater deprived arid and semiarid estuaries from the Northeast Brazil (TA), Senegal (TA), Kenya (WIP) and Iran (WIP) highlight the influence of high temperatures and extreme salinity (hypersalinity) values on the ichthyoplankton assemblages (SLOTERDIJK *et al.,* 2017; CHERMAHINI *et al.,* 2021; BADÚ, LIMA AND PESSANHA, 2022). The main results of these papers evidence the reduction of survival of fish eggs and larvae due to increase mortality, the simplification of the ichthyoplankton assemblages to stress-tolerant estuarine and marine species, with exclusion of freshwater species, and, therefore, the loss of the nursery function of mangroves for early life stages of fish (CYRUS; VIVIER; JERLING, 2010; CYRUS *et al.,* 2011; ARRUDA JÚNIOR *et al.,* 2023). However, although this condition is expected to become more and more constant, studies on this subject are inexpressive in quantity for tropical estuarine systems, as evidenced in this review (N=3).

The large expanse of mangroves along the Brazilian coast influenced by different hydroclimatic characteristics highlights this country as an important model for studying fish eggs and larvae under different environmental conditions (BLABER *et al.*, 1997; CARRASQUILLA – HENAO *et al.*, 2019; ERMGASSEN *et al.*, 2020; LACERDA *et al.*, 2022). The estuarine systems from the Amazonian coast of Brazil are directly influenced by the freshwater discharge of the great Amazon River, which influences the salinity variation, the structure of ichthyoplankton and the nursery function of the mangroves (CONTENTE *et al.*, 2007; COSTA *et al.*, 2011; ZACARDI; SOBRINHO; SILVA, 2014). Due to its importance as a food source and protection from predation, these mangroves are considered spawning zones and as part of the life cycle of economic interest fishes species, such as the sciaenid *Cynoscion acoupa*, found in large abundance as post flexion larvae in the mangroves creeks of Caeté estuary (BARLETTA; SAINT-PAUL, 2002b; MATOS; LUCENA, 2006).

On the other hand, estuaries from the *Northeast and Eastern marine ecoregion* are influenced by two climate types: (I) semiarid climate (*BSh*) and (II) tropical wet and dry (*Af*) (OLIVEIRA; SILVA; LIMA, 2017; BECK *et al.*, 2018; DUBREUIL *et al.*, 2018). The former is distributed along a region known as the Semiarid Coast of Brazil (SCB) and the latter extends along the eastern coast of Brazil (ECB).

Estuaries in the Semiarid Coast of Brazil are classified as shallow, low inflow, and drought prone, due to the influence of the Semiarid climate that is characterized by high temperature values (26–30 °C) and little precipitation or concentrated precipitation in a single month (MARENGO *et al.*, 2016; SOARES *et al.*, 2021). Severe and constant drought events intensify these characteristics and directly increase the salinity of these systems, resulting in a condition known as hypersalinity (TWEEDLEY *et al.*, 2009).

4.2 Environmental drivers of the ichthyoplankton assemblages in mangroves

Among the environmental variables tested, water temperature and salinity are the main factors that determines the structure and density of fish eggs and larvae in the tropical estuaries analyzed. Variations in the abundance and diversity of fish eggs and larvae are linked to the reproductive strategies of adults and to the climatic variables that influence estuarine systems (ARSHAD *et al.*, 2012; GUERRERO *et al.*, 2021). Although temperature does not vary as much in tropical environments, salinity is a factor that varies over space and time at different scales (TELESH; KHLEBOVICH, 2010).

The estuarine distribution of fish larvae in the estuaries analyzed are linked to the salinity gradient of the systems. In the main channel of the Caeté estuary (TA – Amazonian Coast)**, s**pecies of the marine origin were mainly captured in the lower estuary (20 – 40), whereas the upper estuary (0 – 10) consists of taxa associated with freshwater conditions (BARLETTA-BERGAN, BARLETTA, SAINT-PAUL, 2002a). Same result was found for the Mahury estuary (TA – French Guiana, which is influenced by the plume of the Amazon River, where marine species are associated with the lower portion of the estuary (high salinity) and more estuarine and freshwater species are associated with the upper portion (low salinity) (ROUSSEAU *et al*., 2017).

Saline intrusion is more persistent in estuaries from arid and semiarid regions, due to typical short rainy season and little or no freshwater discharge (POTTER *et al.*, 2010; VALENTIM; MENEZES; TEIXEIRA, 2018). An early study in the East African mangroves from Kenya (WIP) considered this tropical estuarine system only a nursery zone for the resident species, possibly due to the ability of these organisms to withstand the high salinity value of the system (at about 35), to the detriment of non-resident species (LITTLE, REAY, GROVES, 1988).

Estuaries in the Northeast Brazil are not influenced by large rivers and salinity plays an important role in structuring the estuarine ichthyoplankton assemblages (BONECKER; CASTRO; BONECKER, 2009; MARCOLIN *et al.*, 2010; SANTOS; RAMOS; BONECKER, 2017a). Salt intrusion in the Vaza Barris estuary extends up to 20 km upstream and reaches values of 30-35 during the dry season and 5-25 during the rainy season (SANTOS; SEVERI, 2018). This results in a decrease in diversity from the river mouth to the upper estuary due to the absence of freshwater species and entrance of marine species that take advantage of the saline intrusion and less freshwater discharge. This pattern of species distribution along estuaries in this region has also been observed in other estuaries, where marine and estuarine species (rarely or never freshwater species) constitute the assemblage of fish egg and larvae in terms of density and diversity (LIMA; COSTA; BARLETTA, 2014; LIMA; COSTA; BARLETTA, 2015; LIMA *et al.*, 2015).

It is important to note the occurrence of studies investigating water quality and pollutants on the structure of fish eggs and larvae (SARPEDONTI; ANUNCIAÇÃO; BORDAÇO, 2013; SANTOS; RAMOS; BONECKER, 2017b) and the interaction between fish larvae and microplastics (LIMA; COSTA; BARLETTA, 2014; LIMA; BARLETTA; COSTA, 2015; LIMA *et al*., 2015).

The bioavailability of pollutants in the estuarine environment enables their insertion into the estuarine trophic web and negatively alters species survival, since fish eggs and larvae are very sensitive (ATHEY *et al.,* 2020; BARLETTA; COSTA; DANTAS, 2020). The presence of microplastic in the estuarine systems ends up making this pollutant available for ingestion by fish larvae, either through the water column or by ingesting contaminated prey, which results in mortality or deficiency in larval development (STEER *et al.,* 2017; RODRIGUES *et al.,* 2019; PANNETIER *et al*., 2020). Some studies also highlight the influence of xenobiotics, oil spills and collapse of dams in the abundance of fish larvae on occurrence of embryonic malformations, which can lead to lower survival of these organisms (OSTRACH *et al.,* 2008; BONECKER *et al*., 2022; SOUSA *et al.,* 2022). In this sense, studies that show the negative influence of human pressure on the nursery function of mangroves are extremely important, as an alert to the importance of spatial planning and protection of mangroves.

Interesting, in Vitória Bay **(**TA **-** Southeastern Brazil) (JOYEUX; PEREIRA; ALMEIDA, 2014) and in Bahía Málaga Estuary (Pacific Coast of Colombia - TEP) (MEDINA-CONTRERAS *et al.,* 2014) did not find any relation of ichthyoplankton assemblage with salinity, temperature or other abiotic variables. The presence of resident or coastal species in these estuaries, with the ability to withstand a wide variation in salinity (euryhaline) and to distribute themselves homogeneously in these estuaries, may have been a key factor in obtaining this result. In addition, other untested variables in these studies, such as zooplankton biomass, primary production, and competition/predation intensity are possible to influence the community.

4.3 The nursery function of mangroves and main taxonomic groups that occur in these systems

Food and shelter are sought for early life states of several species of fish in mangroves, where they grow and develop to latter recruit in the fish stocks (LEFCHECK *et al.*, 2019; VORSATZ, PATTRICK, PORRI, 2021). The studies of ichthyoplankton diversity and distribution of Matang mangrove forest in Malaysia (WIP) reinforce the importance of mangroves as nursery and feeding zones for early life stages of fish (ARA *et al*., 2020). These organisms can take advantage of tidal transport or even travel into the mangroves in search of food, in addition, they can be retained in the mangrove creeks, which prevents them from being transported to less productive areas (OOI; CHONG, 2011; ARA *et al*., 2020).

Estuarine and marine species dominates the mangroves of the tropical estuaries; however, freshwater species are less common, mainly restricted to a few species in the upstream zones (low salinities), during the rainy season or totally absent in arid and semiarid estuaries. The most abundant species in these studies belong to four main families with freshwater, resident, and marine representatives.

Engraulidae are quite common in estuaries, where their eggs and larvae can be found in great abundance throughout the year due to their broadcast spawning events (MCBRIDE *et al.,* 2013). The eggs are ellipsoid and hardly misidentified due to this morphological characteristic (FAVERO *et al.,* 2014). Due to their wide ability to tolerate different salinity levels, they are mainly found in coastal marine environments, but can migrate to freshwater areas (NELSON, 2006). The organisms in this group are an important component of the food chain, since they prey on zooplankton and are abundant prey for piscivorous fishes (ARAÚJO *et al.*, 2008).

Gobiidae (or Gobies) is the most diverse family of amphidromous species adapted to extreme climatic and hydrological seasonal variation, which allows them to habitat different environments (KEITH, 2003). The eggs of this group are demersal, but the larvae are pelagic, preying of microcrustaceans and microalgae in the water column (BORGES, 2011). They are major example of ornamental fishes, due to their diverse coloration and intriguing behaviors, therefore, they are quite common in ornamental fish production companies. (LINDEN *et al.*, 2020).

Clupeidae are a family that together with Engraulidae is part of the order Clupeiformes. This family are the most important fishes economically, as food for both human and many other commercial fish species (COAD, 2017). Fish larvae of this group can be found in freshwater, estuarine and marine environments, some of them are anadromous (ARA *et al.*, 2011). They are widely recognizing as euryhaline and are able to withstand the changes of salinity in mangroves, even found in hypersaline estuaries (BORNMAN; STRYDOM; CLEMMESEN, 2018; GUYAH; WEBBER; PROSPERE, 2021).

Sciaenidae are commonly known as Croakers that are bottom-dwelling carnivore fishes that can tolerate a wide range of salinity and inhabit the coastal and estuarine zones of many regions of the world (MARCENIUK *et al.*, 2019; BARMAN *et al*., 2022). They are worldwide commercially important group of fishes, but they are suffering from population declines due to overfishing (CHAO *et al.,* 2015). Mangroves are important habitat and nursery grounds for this group, so the preservation of these environments is essential for the recovery of these populations and the maintenance of fish stocks (CASTELANNOS – GARLINDO *et al.,* 2012; VILLAR *et al.,* 2013).

4.4 Sampling methods used: what can be improved?

Two issues were raised from the studies analyzed in this review related to the quantification of eggs and larvae and the sampling apparatus. Overall, studies on ichthyoplankton seek to understand distribution patterns of fish eggs and larvae at different spatial scales, through comparison between aquatic systems (STRYDOM, 2015; SANTOS; RAMOS; BONECKER, 2017a). However, it is common to find it difficult to make comparisons between these studies, because, as evidenced, the volume used to calculate the density of organisms is not standardized (10, 50, 100 and 1000m^3). In this sense, with the goal of understanding inter-estuarine ichthyoplankton variations in tropical systems through comparisons, researchers need to standardize the volume they are using for the calculation of average density of organisms.

Regarding the sampling apparatus, the efficiency of different types of nets and the use of different mesh sizes are well documented for Zooplankton (JOHSON; MORSE, 1994; PEPIN; SHEARS, 1997; HERNÁNDEZ-JÚNIOR *et al*., 2011; JOHNSON; FOGARTY, 2013). Different mesh sizes provide more comprehensive information about the size, abundance, and diversity of these organisms (especially for eggs and larvae) (ANTACLINI; HERNÁNDEZ; SABATINI, 2010; MACK *et al*., 2012; PANSERA *et al*., 2014).

Typically, smaller larvae are captured with smaller mesh sizes, but large larvae are captured with larger mesh sizes (SCHOBERND *et al*., 2018). Fish eggs and larvae change in size as they develop, and the appearance of new structures, such as fins, enables the larvae to move in search of food (DOWNIE *et al*., 2020). A better sampling design should capture as many larvae at different developmental stages as possible to understand the use of the mangroves by different species of early life stages of fish, whether as spawning, rearing or feeding zones. Therefore, we encourage the researchers to test the hypothesis that the use of different mesh sizes can capture organisms at different stages of development, as a way of understanding what the function of mangroves is for these organisms.

5 CONCLUSIONS AND FINAL REMARKS

This review elucidated the influence of environmental variables on the structure of fish eggs and larvae in tropical estuarine systems, through studies on this topic. In addition, aspects related to scientific production, sampling methodology and description of the assemblage in the tropical estuaries analyzed were discussed. Mangroves from these tropical systems provide food and shelter for early life stages of fishes, therefore, acting as nursery zones. Despite the importance of this topic, the annual scientific production on ichthyoplankton in these tropical coastal environments is still relatively scarce, which calls attention to the need for further studies.

Research on ichthyoplankton in tropical estuaries is extremely important to understand the importance of mangroves as nursery areas, as well as the major impacts that will follow in these environments as the effects of climate change progress. Therefore, this review not only describes the main characteristics of ichthyoplankton in tropical estuaries but makes important considerations about the new directions that research on this topic should take.

Future studies like the influence of pollutants (e.g microplastics, oil spills, xenobiotics), eutrophication and predation/competition with invasive species in tropical estuarine systems should be conducted since these negative impacts are also present in these systems, impacting the nursery function of mangroves.

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Supplementary material

Table 4 - Selected publications from the bibliographic databases Scopus, Web of science and ScienceDirect used in this review. The respective countries where the studies were conducted, the journals where they were published, and the authors are also presented.

Title of the publication	Country	Journal of publication	Authors
Distribution Gradients of Ichthyoplankton in an East African Mangrove Creek (1988)	Kenya	Estuarine, Coastal and Shelf Science	Little, M.C., Reay, P.J. and Grove, S.J.
The ichthyoplankton of selected estuaries in Sarawak and Sabah: composition, distribution, and habitat affinities (1997).	Indonesia	Estuarine, Coastal and Shelf Science	S. J. M. Blaber, Farmera, M.J., Miltona, D.A., Pang, J., Boon-Teck, O. and Wong, P.
The larval fish fauna of the ‘’Canal de Santa Cruz’’ estuary in Northeast Brazil (2001)	Brazil	Tropical Oceanography	Ekau, W., Westhaus-Ekau, P., Macêdo, S. J., Dorrien,
Structure and Seasonal Dynamics of Larval Fish in the Caeté River Estuary in North Brazil (2002)	Brazil	Estuarine, Coastal and Shelf Science	Barletta-Bergan, A., Barletta, M. and Saint-Paul, U.
Community structure and temporal variability of ichthyoplankton in North Brazilian mangrove creeks (2002)	Brazil	Journal of Fish Biology	Barletta-Bergan, A., Barletta, M. and Saint-Paul, U.

The flood-tide ichthyoplanktonic community at the entrance into a Brazilian tropical estuary (2004)	Brazil	Journal of Plankton research	Joyeux, J.C, Pereira, B. B and Almeida, H. G.
Variação nictemeral do ictioplâncton no estuário do rio Curuçá (Pará-Brasil), durante os períodos chuvoso e seco (2007)	Brazil	Boletim Técnico Científico CEPNOR	Contente, C. T.; Palheta, G. D. A.; Melo, N. F. A. C.; Ramos, C. A. R.; Paiva, R. S.
Larval fish composition of a tropical estuary in northern Brazil (2º18'-2º47'S/044º20'-044º25'W) during the dry season (2007)	Brazil	Pan-American Journal of Aquatic Sciences.	Bonecker, A. C. T.; Castro, M.S.; Namiki, C. A. P.; Bonecker, Barros, F. B. A. G.
Larval fish assemblage in a Tropical estuary in relation to tidal cycles, day/night and seasonal variations (2009)	Brazil	Pan-American Journal of Aquatic Sciences	Bonecker, F.T., Castro, M.S and Bonecker, A.C.T.
Mesozooplankton and Ichthyoplankton composition in two tropical estuaries of Bahia, Brazil (2010)	Brazil	Check list	Marcolin, C.R., Conceição, B.L., Nogueira, M.M., Mafalda Júnior, P. and Johnson, R.
Dynamics of hydrological variables and the fish larva community in an Amazonian estuary of northern Brazil (2011)	Brazil	Journal of Coastal Research	Costa, A. J. G., Costa, K. G., Pereira, L. C. C., Sampaio, M. I. and Costa, R. M.

Influence of temporal and spatial factors on abundance and richness of fish early stages in shallow tropical estuaries (2011)	Mexico	Environmental Biology of Fishes	Arévalo-Frías, W. and Mendoza-Carranza, M.
Larval fish assemblages in a tropical mangrove estuary and adjacent coastal waters: Offshore–inshore flux of marine and estuarine species (2011)	Malaysia	Continental Shelf Research	A.L. Ooi and V.C. Chong.
Larval fish composition and spatio-temporal variation in the estuary of Pendas River, southwestern Johor, Peninsular Malaysia	Malaysia	Coastal Marine Science	Arshad, A.B., Ara, R., Amin, N., Daud, S. K. and Ghaffar, M. A.
Spatio-temporal distribution of fish larvae in relation to ontogeny and water quality in the oligohaline zone of a North Brazilian estuary (2013)	Brazil	Biota Neotropical	Sarpedonti, V., Anunciação, E. M. S. and Bordalo, A. O.
Distribution patterns of microplastics within the plankton of a tropical estuary (2014)	Brazil	Environmental Research	Lima, A. R. A., Costa, M.F. and Barletta, M.

Distribución y densidad de ictioplancton en el Estuario de Bahía Málaga, pacífico colombiano (septiembre de 2009- febrero de 2010) (2014)	Colombia	Boletín de Investigaciones Marinas y Costeras	Medina-Contreras, D., Cantera, J., Escarria, E. and Mejia-Ladino, L.M.
Composição e distribuição de larvas de peixes de um afluente urbano na foz do rio Amazonas, Brasil (2014)	Brazil	Acta of Fisheries and Aquatic Resources	Zacardi, D. M.; Sobrinho, A. F.; Silva, L. M. A. Abdon
Seasonal abundance and distribution of ichthyoplankton diversity in the Coleroon estuarine complex, Southeast coast of India (2014)	India	Biocatalysis and Agricultural Biotechnology	Balakrishnan, T., Sundaramanickam, A., Sudhanshu Shekhar, Muthukumaravel, K and Balasubramanian, T.
Changes in the composition of ichthyoplankton assemblage and plastic debris in mangrove creeks relative to moon phases (2015)	Brazil	Journal of Fish Biology	A. R. A. Lima, M. Barletta, M. F. Costa, J. A. A. Ramos, D. V. Dantas, P. A. M. C. Melo, A. K. S. Justino and G. V. B. Ferreira
Seasonal distribution and interactions between plankton and microplastics in a tropical estuary (2015)	Brazil	Estuarine, Coastal and Shelf Science	Lima, A. R. A., Barletta, M. and Costa, M. F.

O ictioplâncton e sua relação com a variação diária e os ciclos de marés nos estuários amazônicos (2016)	Brazil	Biota Amazônica,	Zacardi, D. M.; Bittencourt, S. C. S.; Nakayama, L.
Can we assess the ecological status of estuaries based on larval fish assemblages? (2017)	Brazil	Marine Pollution Bulletin	Santos, R.V.S., Ramos, S. and Bonecker, A.C.T.
Environmental control on larval stages of fish subject to specific salinity range in tropical estuaries (2017)	Brazil	Regional Studies in Marine Science	Santos, R.V.S., Ramos, S. and Bonecker, A.C.T.
Spatial distribution and seasonality of ichthyoplankton and anthropogenic debris in a river delta in the Caribbean Sea (2017)	Colombia	Journal of Fish Biology	Correa-Herrera, T., Barletta, M., Lima, A.R.A., Jiménez-Segura, L.F. and Arango-Sánchez, L.B.
Composition and structure of the larval fish community related to environmental parameters in a tropical estuary impacted by climate change (2017)	Senegal	Estuarine, Coastal and Shelf Science	Sloterdijk, H., Brehmer, P., Sadio, O., Müller, H., Doring, J. and Ekau, W.
Dynamics of early life-history stages of fish along an estuarine gradient (2008)	Brazil	Fisheries Oceanography	Santos, R. V. S and Severi, W.

Spatiotemporal dynamics of larval fish in a tropical estuarine mangrove: example of the Mahury River Estuary (French Guiana) (2018)	French Guiana	Canadian Journal of Fisheries and Aquatic Sciences	Rousseau, Y., Blanchard, F. and Gardel, A.
Larval fish assemblage, diversity and habitat ecology in the Matang Mangrove Ecosystem, Perak, Malaysia (2020)	Malaysia	Journal of Environmental Biology	Ara, R., Amin, S. M. N., Yusoff, F. M., Arshad, A and Romano, N. R.
Diversity, distribution, and abundance patterns of ichthyoplankton assemblages in some inlets of the northern Persian Gulf (2021)	Iran	Journal of Sea Research	Chermahini, M. A., Shabani, A., Naddafi, R., Ghorbani, R., Rabbaniha, M. and Noorinejad, M.
Environmental influences on the ichthyoplankton in hypersaline estuaries located in a Semiarid Northeastern Brazilian coast (2022)	Brazil	Neotropical Ichthyology	Badú, M. L., Lima, C. S. S. and Pessanha, A. L. M.

CHAPTER 02 – Analysis of a hypersaline drought-prone estuary reveals low density and diversity of fish eggs and larvae (Baseline paper)

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HIGHLIGHTS

- Stress-tolerant species compose the ichthyoplankton of this hypersaline mangrove.
- Most larvae in the pre-flexion stage may be a response to hypersalinity.
- Extreme salinity threatens early life stages of fishes in a shallow estuary.
- Droughts and hypersalinity may affect the nursery function of mangroves.

ABSTRACT

We analyzed fish eggs and larvae in an estuary under severe drought conditions. We detected an inverse salinity gradient, with values increasing from the mouth to the upper estuary. Egg densities decreased from the estuarine mouth to the upstream areas following the salinity increase for all three mesh net sizes. This pattern was also found for the density of larvae, which decreased in estuarine regions with hypersalinity (38 to 62). The low diversity constituted only nine fish species, which were classified as anadromous (*Anchoa hepsetus)*, estuarine and marine (*Bathygobius soporator*, *Hippocampus reidi*, *Eucinostomus* sp., and *Diapterus auratus*), marine estuarine opportunist (*Caranx latus*

and *Bardiella rochus*), and marine stragglers (*Echeneis naucrates* and *Haemulon* sp.). In addition, we observed an oversimplification of the assemblage to include stress-tolerant estuarine and marine species. Our baseline results suggest that this hypersaline estuarine ecosystem has lower densities and diversity than a healthy mangrove system.

GRAPHICAL ABSTRACT

1 INTRODUCTION

Estuaries are key coastal ecosystems for many species of fish worldwide because of their ecological functions as feeding, spawning, and nursery habitats (RAMOS *et al*., 2012; POTTER *et al*., 2015; WHITFIELD, 2016, WHITFIELD, 2017; KISTEN AND STRYDOM 2021; GUERREIRO *et al*., 2021). Fish eggs and larvae, named ichthyoplankton, drift in the water column of these estuarine systems and interact with prey, predators, and a range of environmental variables (HOUDE, 2001; ARÉVALO-FRIAS AND MENDONZA-CARRANZA, 2015; ZHANG et al., 2022). In this regard, studies on these organisms and their dynamics are of great relevance to fisheries management, especially in low-inflow or freshwater-deprived mangrove estuarine ecosystems. As the early life stages of fish are more vulnerable to mortality, the effects of extreme events in estuaries is a determinant of recruitment (CABRAL *et al*., 2021; COLOMBANO *et al*., 2022).

Tropical estuaries in arid and semiarid regions are characterized by low freshwater inflow and high rates of evaporation, some of which are intermittently blocked from the sea by sand spits (POTTER *et al*., 2010; TWEDLEY *et al*., 2019). The combination of these factors and human intervention can result in hypersalinization, which means that the salinity of the estuary is higher than that of the adjacent ocean (ANDUTTA *et al.*, 2011). This extreme phenomenon is especially important during the dry season when salinity increases. Hypersaline conditions expose aquatic biota, such as fish eggs and larvae, to osmotic stress and metabolic changes that affect their temporal and spatial distribution as well as survival rates (Tweedley et al., 2019; Whitfield et al., 2006). Furthermore, increased salinity is an important factor that can simplify the estuarine communities, select stress-tolerant species (Barroso et al., 2018), and reduce the overall abundance and diversity of organisms (CYRUS *et al.*, 2010; CARRASCO AND PERISSINOTTO, 2012; ROSA *et al.*, 2016; SLOTERDIJK *et al.*, 2017).

The intensification of climate change effects, such as sea level rise, reduced precipitation, and high levels of evaporation (CAI *et al.*, 2022) results in the hypersalinization of estuaries. On the Brazilian semiarid coast, short estuaries are already experiencing hypersalinity due to local impacts (e.g., multiple dams) and ongoing climate change, which makes them useful models for understanding this phenomenon (SCHETTINI *et al.*, 2017; SOARES *et al.*, 2021). However, few studies have analyzed these extreme mangroves and their ichthyoplankton communities (BADU, LIMA, PESSANHA, 2022). Using a semiarid estuary in an extreme drought year as a model, we analyzed the intra-annual distribution and density of fish eggs and larvae.

2 MATERIAL AND METHODS

2.1 Study site

The Piranji River Estuary (Semiarid Coast of Brazil) is located on the Ceará state coast in the Equatorial Southwestern Atlantic. The study region is located in a small, shallow hydrographic basin, 55 km long and 4.367 km2 total area (SILVA; SILVA, 2012) (Figure 1). This low-inflow estuary is located on the Brazilian semiarid coast, and its morphology is composed of narrow, shallow channels with extensive sand and mud banks. Moreover, there is a 3.2 km long sand spit at the mouth (SILVA *et al.*, 2012) that decreases the tidal flow inside the estuary.

The rainy period lasts from January to June, and the dry period from July to December (LACERDA *et al.*, 2007) (Figure 2). This mangrove estuarine ecosystem can experience hypersaline conditions because of several factors, such as a semiarid climate, drought-prone area, high evaporation rate, high water residence time, and low levels of surface runoff (SCHETTINI *et al.*, 2017).

Figure 1 – Sampling points in the Piranji River Estuary, Northeast Brazil, P1, upstream; P2, intermediate; and P3, downstream, near the estuarine mouth and sand spit.

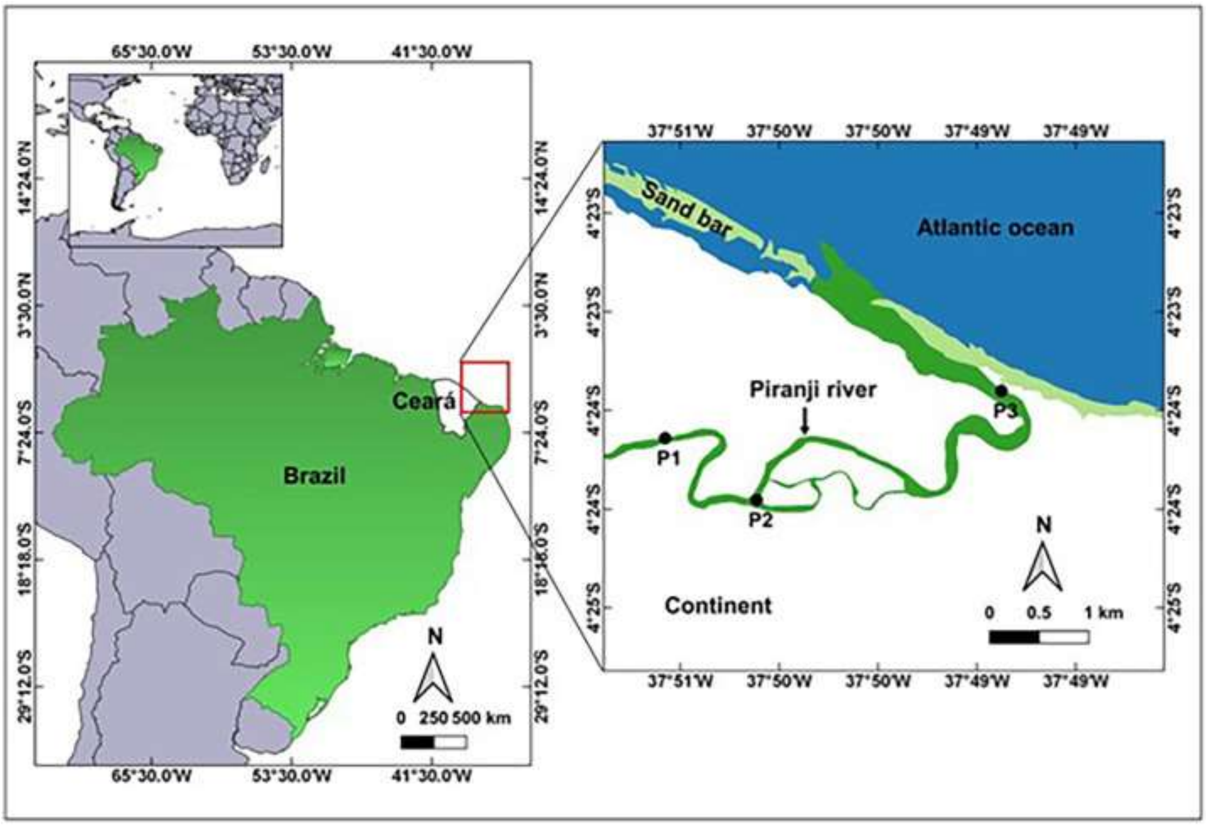
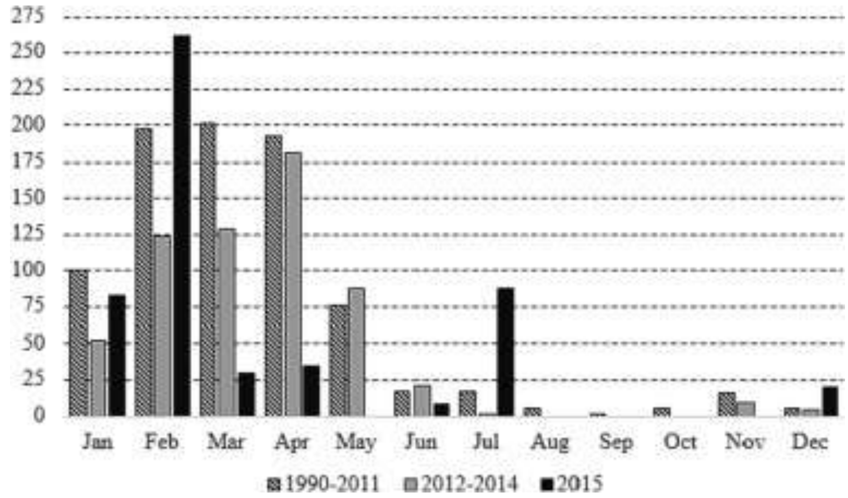


Figure 2 – Mean historical precipitation (1990–2011), precipitation during a historic drought (2012–2014), and precipitation during the study period (2015), Northeast Brazil.



Natural drought cycles are characteristic of this semiarid region and have increased between 2000 and 2020, correlated with El Niño conditions and the positive phase of the Atlantic Meridional Mode (MARENGO *et al.*, 2017). The combination of both mechanisms from 2010 to 2016 led to the most severe drought (Figure 2) ever recorded in this region (MARENGO *et al.*, 2017).

2.2 Sampling design

Intra-annual sampling occurred in 2015 during the diurnal period in the rainy (February, April, and June) and dry seasons (August, October, and December). Data were collected at three different points, P1 (upstream river station), P2 (intermediate station), and P3 (downstream river station, near the river mouth), along the Piranji Estuary (Figure 1) during the ebb tide. The depth, water temperature, salinity, and dissolved oxygen levels were measured at each station using a YSI 6600 multiparameter meter.

Horizontal surface hauls for ichthyoplankton samples were collected by boat at all three stations using conical nets of 200, 300, and 500 μm mesh (mouth diameter: 50 cm) equipped with a flowmeter (General Oceanics, Miami, FL, USA). The nets were towed at approximately 2 knots for 3 min at each station. All samples were immediately fixed in 4 % formalin buffered with borax.

2.3 Data Analysis

Ichthyoplankton density is expressed as the number of individuals/100m³ (OMORI; IKEDA, 1984). Only the eggs of the Engraulidae family were identified and all others were classified as morphotypes A, B, C, D, and E, according to their morphometric (egg size) and morphological characteristics. Fish larvae were identified to the lowest taxonomic level (according to, (RÉ, 1999; RICHARDS, 2005) and classified into different stages of development, according to the flexion of the notochord, as pre-flexion, flexion, and post-flexion, based on RÉ (1999). Ecological guild classification considers the habits of breeding adults and their migration patterns. The following characteristics were used: location of spawning, feeding, and/or refuge, which in some cases involve migratory movements between estuaries and other ecosystems (ANDRADE-TURBINO *et al.*, 2008; POTTER *et al.*, 2015).

3 RESULTS

3.1 Environmental variables

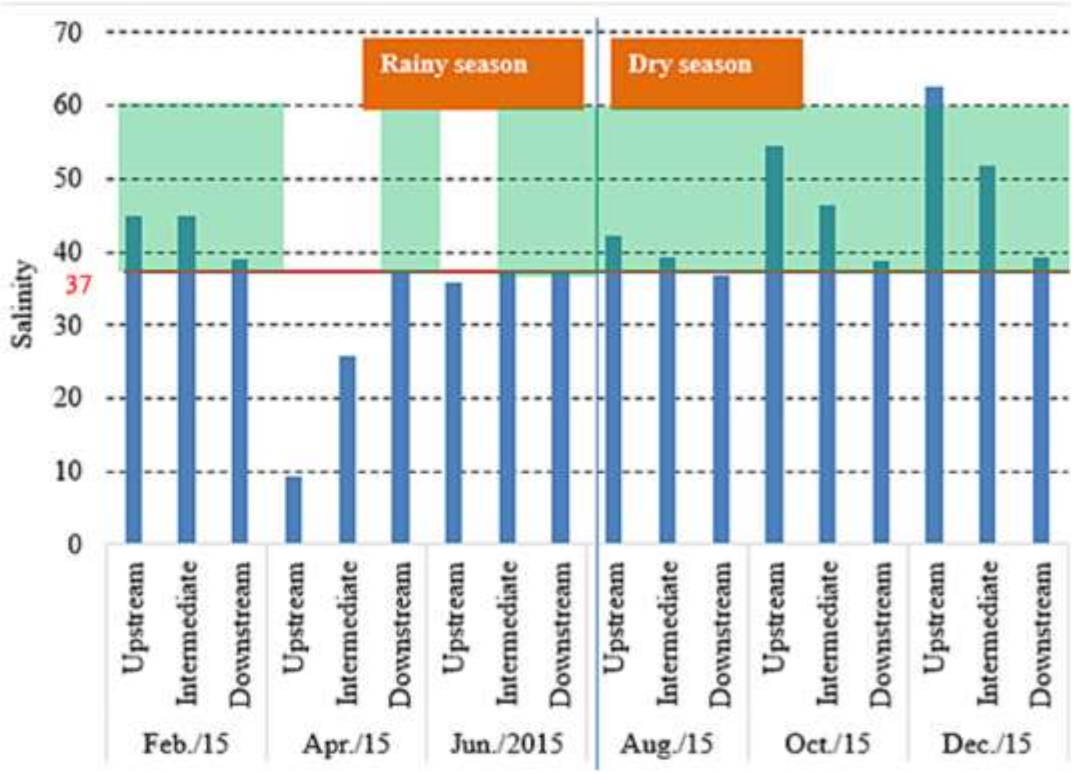
The estuary depth was typical of shallow, low-inflow, short estuaries, deeper at P3, the downstream station, near the river mouth, during the rainy (4.95 ± 0.67 m) and dry seasons (3.65 ± 0.28 m), and shallower at P2, the intermediate station (2.46 ± 0.18 m), and at P1, the upstream station (2.3 ± 0.37 m), during the rainy season and the dry season, respectively (Table 1). The water temperature was nearly constant during the study, as expected for low-latitude estuarine systems, at 28.4 ± 0.36 (P1), 28.2 ± 0.16 (Intermediate) and 27.66 ± 0.32 (Downstream), during the rainy season, and 27.55 ± 0.43 (Upstream), 26.82 ± 0.68 (P2) and 26.12 ± 0.44 (Downstream), during the dry season (Table 1).

Table 1 – Physical-chemical variables measured in the Piranji Estuary, Northeast Brazil, during the rainy and dry seasons.

	Rainy Season			
	Depth (m)	**Temperature (ºC)**	**Salinity**	**Dissolved Oxygen (mg/L)**
Upstream	3.19 ± 0.65	28.4 ± 0.36	30.1 ± 15.1	5.43 ± 0.32
Intermediate	2.46 ± 0.18	28.2 ± 0.16	36 ± 7.83	5.33 ± 0.36
Downstream	4.95 ± 0.67	27.66 ± 0.32	**38 ± 0.71**	6 ± 0.43
	Dry Season			
	Depth (m)	**Temperature (ºC)**	**Salinity**	**Dissolved Oxygen (mg/L)**
Upstream	2.3 ± 0.37	27.55 ± 0.43	**53.2 ± 8.38**	5.41 ± 0.5
Intermediate	3.03 ± 0.91	26.82 ± 0.69	45.76 ± 5.12	5.59 ± 0.21
Downstream	3.65 ± 0.28	26.12 ± 0.44	38.4 ± 1.08	6.01 ± 0.01

High levels of dissolved oxygen were found at Downstream during the rainy (6 ± 0.43 mg/L) and dry seasons (6.01 ± 0.01 mg/L), but these decreased at Upstream and Intermediate during the rainy (5.33 ± 0.36 and 5.43 ± 0.32 mg/L) and dry seasons (5.59 ± 0.21 and 5.41 ± 0.5 mg/L) (Table 1). During the rainy season, salinity levels were high at all three sampling stations, but only Downstream exceeded the salinity of the adjacent sea. During this period, the salinity was highest downstream at Downstream (38 ± 0,71), followed by Intermediate (36 ± 7.83), and lowest at Upstream (30.1 ± 15.1). In the dry season, the salinity changed drastically; it was lowest at Downstream near the ocean (38.4 ± 1.08), followed by Intermediate (45.76 ± 5.12), and highest at Upstream in the uppermost estuarine area (53.2 ± 8.38) (Table 1). Although the station closest to the sea had the highest salinity during the rainy season, in the dry period, this pattern was reversed and the estuary became hypersaline, with salinity levels increasing upstream (Figure 3, green rectangle).

Figure 3 – Salinity levels measured at three sampling stations during the rainy and dry seasons in the Piranji Estuary, Northeast Brazil. The solid red line represents the salinity of the adjacent sea, the green rectangle represents the stations and months with hypersalinity. P1, upstream; P2, intermediate; P3, downstream.



3.2 Density of fish eggs and larvae and stages of development

The total numbers of fish eggs were 189, 100, and 174 in the 200, 300, and 500 µm nets, respectively. Only Engraulidae and five morphotypes (A, B, C, D, and E) comprised the estuarine egg assemblage. There was a decrease in egg density for all three mesh net sizes from the mouth to the upper estuary, i.e., downstream to upstream (Table 2). The highest egg densities were observed at Downstream (P3), near the ocean (Table 2), where the total density was 124.9, 73, and 40.7 eggs/100m3 in the 200, 300, and 500 µm net, respectively. The lowest densities were observed at Upstream (P1) (Table 2) with 2.8, 0.8, and 0.5 eggs/100m3 in the 200, 300, and 500 µm nets, respectively.

Table 2 – Morphotypes of fish eggs found in the Piranji Estuary, Northeast Brazil, and their respective densities at each sampling station and net mesh size.

	Net mesh size (µm)	**200**			**300**			**500**		
	Station	**P1**	**P2**	**P3**	**P1**	**P2**	**P3**	**P1**	**P2**	**P3**
Density (eggs/100m^3)	**Engraulidae**	2.8	1.4	56.2	0	0	30.1	0	0	11.1
	Morphotype A	0	0	18.7	0.8	0	0	0	0	5.3
	Morphotype B	0	0	29.1	0	0	25.6	0	0	12.5
	Morphotype C	0	2.1	20.8	0	1.5	14.3	0	0	1.2
	Morphotype D	0	0	0	0	0	3.0	0	0	0.7
	Morphotype E	0	0	0	0	0	0	0	0	9.9
	Total	**2.8**	**3.5**	**124.9**	**0.8**	**1.5**	**73.0**	**0.5**	**0.7**	**40.7**
	Pattern of distribution	**P1<P2<P3**			**P1<P2<P3**			**P1<P2<P3**		

The total numbers of fish larvae were 183, 96, and 69 in the 200, 300, and 500 µm nets, respectively. The lower fish larval diversity was composed of nine taxa distributed across eight families, nine genera, and nine species (Table 3). Considering each mesh size, the total density of fish larvae increased towards the river mouth, except for the 200 µm net when the density was higher at P1 than P2 (Table 3). The highest total densities of fish larvae were observed at P3 with 90.9, 36.1, and 9.9 larvae/100m3 collected in the

200, 300, and 500 µm nets, respectively. Conversely, the lowest total densities of fish larvae were observed at P2 (15.3 larvae/100m³) with the 200 µm net, and at P1 (19.6, and 3.1 larvae/100m³) in the 300, and 500 µm nets, respectively (Table 3).

Anchoa hepsetus was found at the highest density (mean = 16.78 larvae/100 m3) among all the organisms collected with the 200, 300, and 500 µm nets at the three sampling stations. *Bathygobius soporator* (mean = 4.0 larvae/ 100 m3), *Eucinostomus* sp. (mean = 1.74 larvae/100 m3), and *Haemulon* sp. (mean = 1.0 larvae/100 m3) were the other most representative taxa in the samples. The assemblage of fish larvae was composed almost entirely of taxa belonging to estuarine and marine guilds (Table 3). We recorded all three developmental stages at each sampling station, with a predominance of larvae in the pre-flexion stage (18.39, 17.53, and 59.48 larvae/100m3 at P1, P2, and P3, respectively) (Fig. 4). The flexion stage was only recorded at P3 (Fig. 4).

Figure 4 – Larval developmental stages found at the sampling stations along Piranji estuary, Northeast Brazil. PF, Preflexion; F, Flexion; POF, Postflexion. P1, upstream; P2, intermediate; P3, downstream.

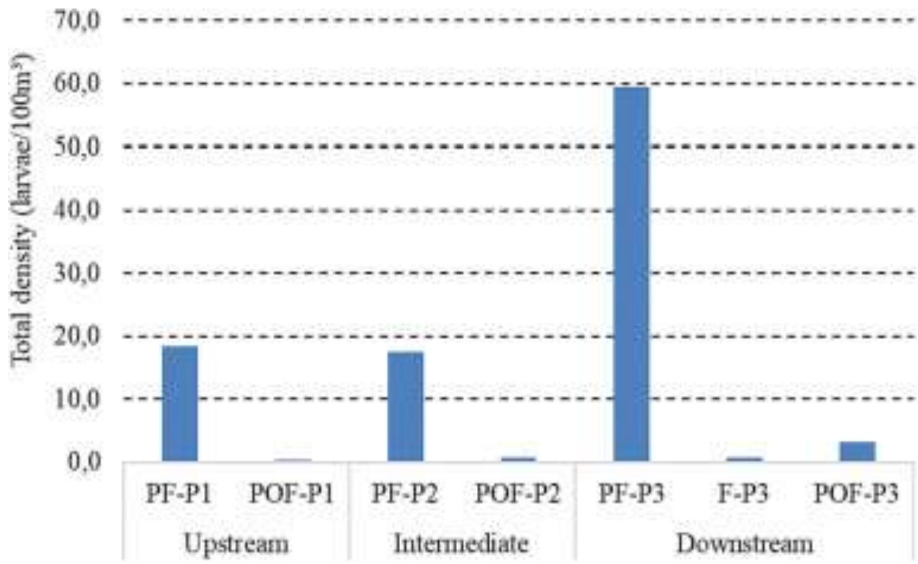


Table 3 - Taxa of fish larvae found in the Piranji estuary, Northeast Brazil, and their respective densities at each sampling station and with each net size, and their classification according to ecological guild. P1, upstream; P2, intermediate; P3, downstream.

Family	**Species**	**Mesh size (200 µm)**			**Mesh size (300 µm)**			**Mesh size (500 µm)**			**Ecological Guilds**
		P1	**P2**	**P3**	**P1**	**P2**	**P3**	**P1**	**P2**	**P3**	
Gobiidae	*Bathygobius soporator* (Fillfrin Goby)	0	1.39	9.02	0	9.79	8.28	1.68	0	2.89	Estuarine and Marine
Engraulidae	*Anchoa hepsetus* (Broad-striped Anchovy)	15.27	12.49	71.48	16.57	9.79	20.33	1.20	1.20	2.65	Anadromous
Gerreidae	*Eucinostomus* sp. (Mojarra)	5.55	1.39	0	0	0	7.53	0	0.72	0.48	Estuarine and Marine
	Diapterus auratus (Irish Mojarra)	0	0	0	0	0	0	0	0	1.20	Estuarine and Marine

Haemulidae	*Haemulon* sp. (Scaled fin Grunts)	0	0	10.41	0	0	0	0	0	0.72	Marine stragglers
Carangidae	*Caranx latus* (Horse-eye Jack)	0	0	0	0	0	0	0.24	0.48	0.24	Marine estuarine - opportunist
Echeneidae	*Echeneis naucrates* (Live Shark-sucker)	0	0	0	0	0	0	0	0	0.48	Marine stragglers
Sciaenidae	*Bairdiella ronchus* (Ground Croaker)	0	0	0	0	0	0	0	0	0.72	Marine estuarine - opportunist
Syngnathidae	*Hippocampus reidi* (Longsnout seahorse)	0	0	0	0	0	0	0	1.20	0.48	Estuarine and Marine
Total density (Larvae/100m³)		**20.8**	**15.3**	**90.9**	**16.6**	**19.6**	**36.1**	**3.1**	**3.6**	**9.9**	
Pattern of distribution		**P1 > P2 < P3**			**P1 < P2 < P3**			**P1 < P2 < P3**			

4 DISCUSSION

Our baseline research evaluated the intra-annual spatial variation in fish larvae and eggs in mangroves under extreme salinity conditions. Environmental variables, such as temperature and dissolved oxygen, in the study area (BARROSO *et al.*, 2018) were similar to those of other tropical estuaries near the equator (MACEDO SILVA *et al.*, 2015; ANDRADE *et al.*, 2016; BASTOS *et al.*, 2016). However, there were remarkable seasonal differences in salinity in two opposing manners (BARROSO *et al.*, 2018): hyposaline and hypersaline regimes. In years of extreme drought, the combined effect of reduced annual rainfall, low water depth, and high evaporation rates promoted hypersalinity in this semiarid estuary (SCHETTINI *et al.*, 2017), even in the rainy season (e.g., February). The El Niño event in 2015 and 2016 was one of the most severe since 1950 (ROSSI; SOARES 2017), and this phenomenon can result in prolonged periods of drought and hypersalinity in mangroves.

Our assessment of ichthyoplankton density and diversity in this hypersaline estuary was unusually low compared to other estuaries without extreme salinity conditions (BALAKRISHNA *et al.*, 2015; ZACARDI, 2015; CORREA- HERRERA *et al*., 2017; Da SILVA *et al.*, 20 *et al.*, 2020; ROUSSEAU *et al.*, 2017). Fish eggs and larvae are even more susceptible to variations in salinity because they cannot osmoregulate like adults, which affects the life history of the early life stages of fish (PÉREZ-ROBLES *et al.*, 2012; ROSA *et al*., 2016; SANTOS *et al.*, 2017).

Salinity is one of the most important factors in ichthyoplankton assemblage distribution and diversity (JAGADEESAN *et al.*, 2013; MARINA *et al.*, 2021). Extreme salinities require high energy rates for osmotic regulation; therefore, the energy that would otherwise be used for the growth and development of eggs and larvae is reduced (KURBEL, 2008). Hypersaline mangroves can impact the life cycle of fishes, including reproduction and alterations in the food web (e.g., ichthyoplankton prey), as well as causing the direct mortality of fish (WHITIFIELD, 2021), which may be among the causes of the low density and diversity recorded in this study and the predominance of larvae in the first stage of development.

The density of fish eggs and larvae are directly influenced by the presence of favorable environmental conditions for adult spawning (SCHRECK *et al.*, 2001; SERVIL *et al.*, 2020). One peculiar characteristic that may have increased the salinity is the presence of a sand spit at the mouth of the estuarine-laguna system. The presence of spits in mangrove estuarine systems reduces their connectivity with the adjacent sea, blocks the entry of breeding adults, fish eggs, and larvae from coastal waters into the estuary, and negatively influences the density and diversity of ichthyoplankton (STRYDOM *et al.*, 2003; WHITFIELD *et al.*, 2006; JAMES *et al.*, 2007; KORSMAN *et al.*, 2017). Other estuarine systems are influenced by the presence of sand spits at their mouths; however, because of the intensifying effects of climate change (e.g., prolonged droughts and river silting), this condition has become more frequent, which prevents the renewal of estuarine water and results in widespread hypersalinity (WHITFIELD, 1992; MORAIS; PINHEIRO, 2011; DESCROIX *et al*., 2020).

Although the fish assemblage from this low-inflow estuary is composed of taxa commonly found in tropical estuarine systems (BONECKER *et al.*, 2009; MARCOLIN *et al.*, 2010; LIMA *et al.*, 2015), the number of species recorded here is considerably low, which is expected in hypersaline coastal environments (ROSA *et al*., 2016; CHERMAHINI *et al.*, 2021). According to POTTER *et al.* (2015), these fish species are classified as anadromous (*Anchoa hepsetus)*, estuarine and marine (*Bathygobius soporator*, *Hippocampus reidi*, *Eucinostomus* sp., and *Diapterus auratus*), marine estuarine opportunists (*Caranx latus* and *Bardiella rochus*), and marine stragglers (*Echeneis naucrates* and *Haemulon* sp.). The lack of freshwater species due to high salinity levels and the occurrence of only marine and estuarine species is a pattern found in other hypersaline aquatic environments (ROSA *et al.*, 2016; BADÚ, LIMA, PESSANHA, 2022), and points to an oversimplification of the fish assemblage under the influence of hypersalinity and severe drought. In addition, the mangrove fish community was composed of salinity-tolerant species.

Bathygobius soporator (Gobiidae) and *Anchoa hepsetus* (Engraulidae) are euryhaline species, which means they can tolerate a wide range of salinities and occupy a great diversity of habitats (NIZINSKI; MUNROE, 2002; SARPEDONTI *et al.*, 2013; MANGAS *et al.*, 2014; SÁNCHEZ-RAMÍREZ; OCAÑA, 2015; MERIGOT *et al.*, 2017), including mangroves experiencing extreme salinity. *B. soporator* eggs are demersal, and their pelagic larvae use estuarine systems to complete their development (JOYEUX *et al.*, 2004). *A. hepsetus* spawns continuously (iteroparity), which explains the high catch of eggs and larvae during ichthyoplankton surveys in estuaries (NIZINSKI; MUNROE, 2002; FAVERO *et al.*, 2015).

Another estuarine and marine larva is the near-threatened *Hippocampus reidi* (Syngnathidae) (ROSA *et al.*, 2007) that was found at the low densities of 1.2 and 0.48 larvae/100 m3 at P2 and P3, respectively, during our study. This species occurs in habitats such as macroalgal beds and mangrove roots (MAI; ROSA, 2009). Salinity experiments with the seahorse *H. reidi* indicates that they better survive and grow at intermediate salinity levels (10–25); thus, the stress caused by hypersalinity should be considered another factor that threatens their population (HORA *et al*., 2016) in mangroves during extreme droughts, as recorded here.

Despite inhabiting marine regions, the family Gerreidae, commonly known as mojarras or silverbiddies, seeks tropical estuaries and mangroves for feeding and reproduction (DE LA CRUZ-AGUERO *et al.*, 2012; RAMOS *et al*., 2016). Many species of gerreids are found from the upper to the lower zones of estuarine systems because of their broad salinity tolerance (0–35) (FRANCO *et al.*, 2012); moreover, they constitute an important resource for the fishing community (RAMOS *et al.*, 2016). In this study, the Gerreidae family was represented by the estuarine and marine species, *Eucinostomus* sp. and *Diapterus auratus*.

Caranx latus (Carangidae) and *Bardiella rochus* (Sciaenidae) are marine estuarine-opportunistic species that enter estuaries to complete their life cycle and then return to the sea (VENDEL; CHAVES, 1998; GUAZZELLI *et al.*, 2021). In contrast, *Echeneis naucrates* (Echeneidae) and *Haemulon* sp. (Haemulidae) larvae are classified as marine stragglers, meaning they enter estuaries sporadically, most commonly where salinity typically does not decline far below 35 (POTTER *et al.,* 2015).

The fish assemblage was dominated by larvae in the pre-flexion stage, with a low density of flexion and post-flexion stages. This can be indicative of spawning activity in the mangrove area, as well as the retention of these organisms in the estuary (KATSURAGAWA; MATSUURA, 1992). Larvae in the pre-flexion stage have little or no swimming ability because their natatory structures are not yet formed, so they are threatened by hypersaline conditions, which cause increased stress (FISHER *et al*., 2000). In addition, larvae do not yet have osmoregulatory organs, such as gills or kidneys; therefore, they are more vulnerable to extreme salinity as more energy is required to maintain osmoregulation (SAMPAIO; BIANCHINI, 2002; OLIVEIRA; PESSANHA, 2014).

Thus, even though the semiarid mangrove in this study is an important nursery for marine fish, the lack of freshwater species and the low densities of larvae in more advanced stages may be indicative of an ongoing loss of its nursery function. This topic requires further long-term study, as previously reported in other regions, for example, the loss of nursery function for fish in Lake St. Lucia Estuary in South Africa. This lake is affected by drought and extended periods of river mouth closure, preventing the entrance of post-larvae into the estuary and the recruitment of juveniles into marine fish stocks (CYRUS. VIVIER, 2006; JAMES *et al*., 2007; CYRUS *et al.*, 2011).

5 CONCLUSION AND FINAL REMARKS

We found a low density and diversity of fish eggs and larvae in this mangrove system during extreme salinity. We also recorded a low number of species adapted to these extreme environmental conditions in the drought-prone mangrove ecosystem. A long time series is needed to better understand the effect of hypersalinity on ichthyoplankton assemblages in extreme estuarine environments. In this context, our results provide an important baseline for long-term ecological analyses on the loss of ecosystem functions, such as mangrove nurseries. Considering the ecological role of ichthyoplankton in mangroves and their vulnerability to global change, our study can help to better understand how this assemblage will respond to future extreme salinity due to drought and warming, with implications for fisheries and mangrove conservation.

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CHAPTER 3 - Composition and estuarine distribution patterns of early life stages of tropical fish under seasonal precipitation

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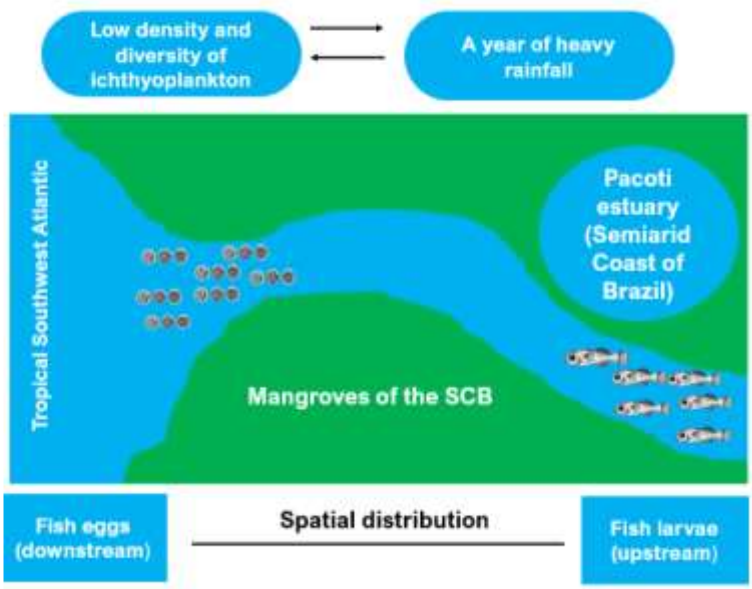
HIGHLIGHTS

- Fish eggs were more abundant downstream, while larvae were mainly abundant upstream.
- Postflexion larvae concentrated upstream evidence the use of this zone as a possible feeding area.
- The assemblage is composed of resident and marine species, with absence of freshwater species.
- In the absence or heavy rainfall, the density of eggs and larvae is reduced
- Intermediate conditions between the dry and rainy periods have higher densities.

GRAPHICAL ABSTRACT



ABSTRACT

The composition and distribution of fish eggs and larvae in tropical estuaries provide science-based information for fisheries resources management and conservation policies. In the Semiarid coasts worldwide, this dynamic is influenced by the extreme seasonality of rainfall. Despite the importance of this research on semiarid estuaries as a model to understand the influence of extreme precipitation events on the nursery function of mangroves for fish, surveys are still scarce in these habitats. Therefore, this study analyzed the structure and distribution of fish eggs and larvae in one tropical semiarid estuary, during extreme precipitation regimes (rainy, transition, and dry season). Five morphotypes and the Engraulidae eggs were mainly concentrated in the downstream in all seasons, with high total densities obtained during the transition season (67.35 eggs/100m³ - 120 µm and 62.65 eggs/100m³ - 300 µm). In addition, during this season, fish eggs were also found in low densities in the intermediate zone. Concerning fish larvae, both 120 and 300 µm nets evidence high densities of fish larvae in the upstream, during all the three precipitation regimes, with a maximum density value of 38.2 larvae/100m³ (120 µm) during the transition season. The fish larvae assemblage was composed by solely estuarine (*Atherinella brasiliensis*, *Gobionellus oceanicus* and *Sphoeroides testudineus.*), marine estuarine-opportunist (*Strongylura* sp. and *Hyporhamphus unifasciatus*) and marine estuarine-dependent species (*Eucinostomus* sp., Sciaenidae sp1. and *Anchovia clupeoides*). Freshwater taxa are absent. Postflexion larvae dominate the upstream zone where high densities of fish larvae from all the ecological guilds were found, which may indicate a possible use of this area for feeding. Overall, the density of fish eggs and larvae were low during the rainy and dry season, in comparison with high densities in the transition season. This baseline assessment provides information on the influence of extreme weather events on the nursery function of tropical estuaries.

Keywords: Fish eggs and larvae; Rainfall; Nursery zones; Salinity; Semiarid Coast of Brazil.

1 INTRODUCTION

Estuaries are transition zones between the mainland and the marine environment, where seawater from the tidal regimes mixes with freshwater discharge (PRITCHARD, 1967;

KENISH, 2002; OLISAH; ADAMS, 2021). Mangroves in tropical estuaries enrich the biological component and act as unique habitats providing food for juveniles and refuge areas (BLABER, 2013; ARCEO-CARRANZA *et al.*, 2021). Among the faunal components, early life stages of fish, ranging from freshwater, estuarine and marine species, take advantage of these seascapes until they recruit and become part of the adult fish stocks (SHEAVES *et al.*, 2015). Hence, tropical estuaries are valuable nursery zones for fisheries worldwide (BLABER, 2013; BABLER; ABLE; COWLEY, 2022).

The composition, abundance and distribution patterns of fish eggs and larvae (ichthyoplankton) are influenced by environmental variables, hydrodynamic forcing, and spawning dynamics of adults (MARTINHO *et al.*, 2012; GUERRERO *et al.*, 2021). Fish eggs and larvae are sensitive life stages, since their body systems are still developing, and therefore are greatly affected by water column variations (PANKHURST; MUNDAY, 2011). The dynamic estuarine environment challenges these organisms, since abiotic variables (e.g., temperature, turbidity, dissolved oxygen) oscillate into the ecosystem on different scales of time and space, influenced by tidal regimes, seasons, and large-scale climatic phenomena (e.g., El Niño and La Niña) (BELARMINO *et al.*, 2021).

In this sense, even though estuarine systems are well known as nursery zones for several species of early life stages of fish, this function is affected positively or negatively by environmental changes (natural or induced) that these systems undergo (VASCONCELOS *et al.*, 2011; MARTINHO *et al.*, 2012; SWADLING *et al.*, 2022) such as extreme precipitation events. Indeed, recent data from the Intergovernmental Panel on Climate Change (IPCC) shows that extreme weather events are becoming more constant worldwide (GEMEDA; KORECHA; GAREDEW, 2022; HALIK; PUTRA; WIYONO, 2022).

Studies on ichthyoplankton in tropical estuaries are still relatively scarce when compared to temperate systems, which hinders the knowledge on the resistance and resilience of these organisms to climate change (ZHANG *et al.*, 2022). This is particularly true for the Semiarid Coast of Brazil, where estuaries (LIMA, PESSANHA, 2022; ARRUDA JÚNIOR *et al.*, 2023) and adjacent coastal waters (MOTA *et al.*, 2014; MOTA *et al.*, 2017; COSTA *et al.*, 2020) are used by early life stages of fish, but still poorly studied in terms of ichthyoplankton ecology. In this context, spatial and temporal variation in semiarid estuaries can provide important insights in the context of global climate change for conservation purposes of mangroves and these tropical systems.

On that basis, we aim to analyze the intra-annual influence of different precipitation regimes (rainy, transition and dry) in the distribution and composition of fish eggs and larvae in one tropical semiarid estuary (northeastern Brazil).

2 MATERIAL AND METHODS

2.1 Study site

The study was conducted in the tropical estuary of Pacoti river, at the Semiarid Coast of Brazil (Northeast Brazil) (Figure 1) (*BSh*, Köppen-Geiger climate system). The precipitation of this region is influenced by N-S Atlantic trade winds that converge along the Intertropical Convergence Zone (ITCZ) and brings rainfall between February - June, followed by a long period of drought (July to January) (BARROS *et al.*, 2019; MONTEIRO; ZANELLA, 2019; MARENGO *et al.*, 2016).

The mean wind speed values are always higher in the second half of the year (Figure 2). Regarding geomorphological and hydrological features, the Pacoti estuary is a short (< 20 km long) and shallow (< 5 m deep) system with semidiurnal tides (ranging between 1.0 and 2.5 m) and ebb-dominant currents (SCHETTINI *et al.*, 2017). During the dry season, the hypersalinization can occur in the system (SCHETTINI *et al.*, 2017).

Figure 1 – Localization of the Tropical estuary of Pacoti river at the Brazilian Semiarid Coast (Southwest Atlantic coast) in the state of Ceará.

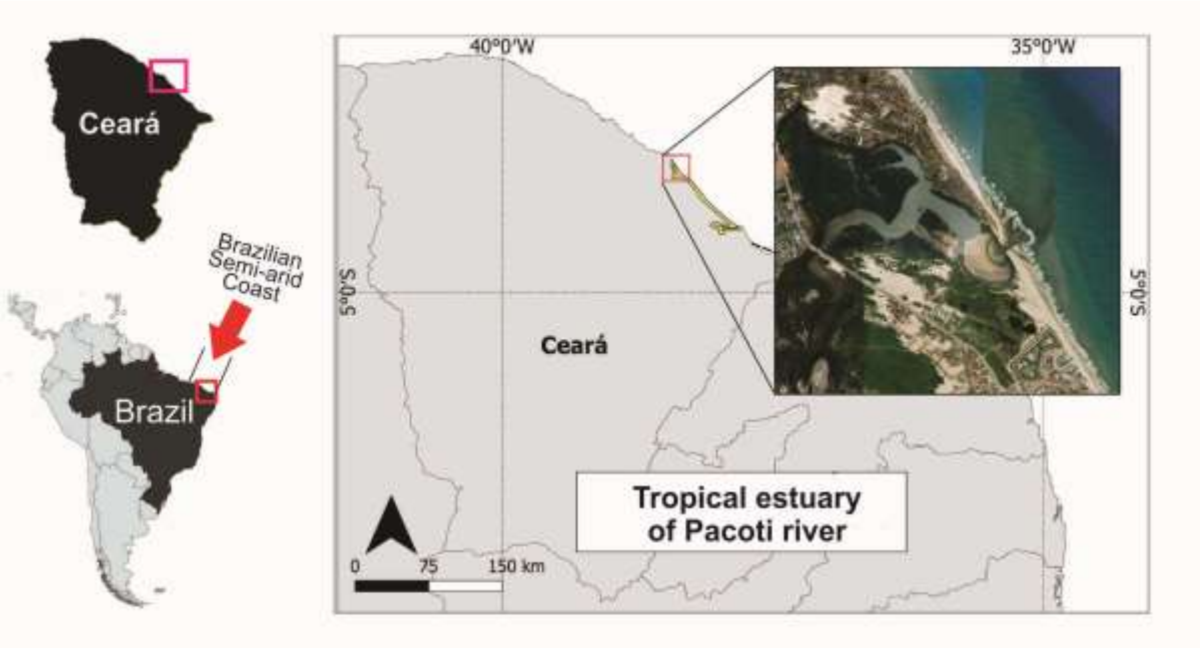
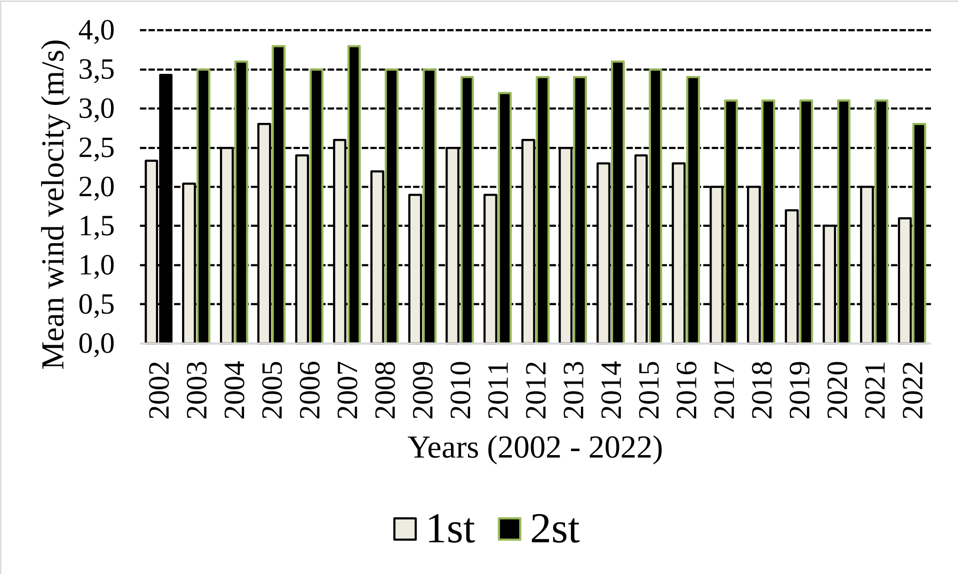
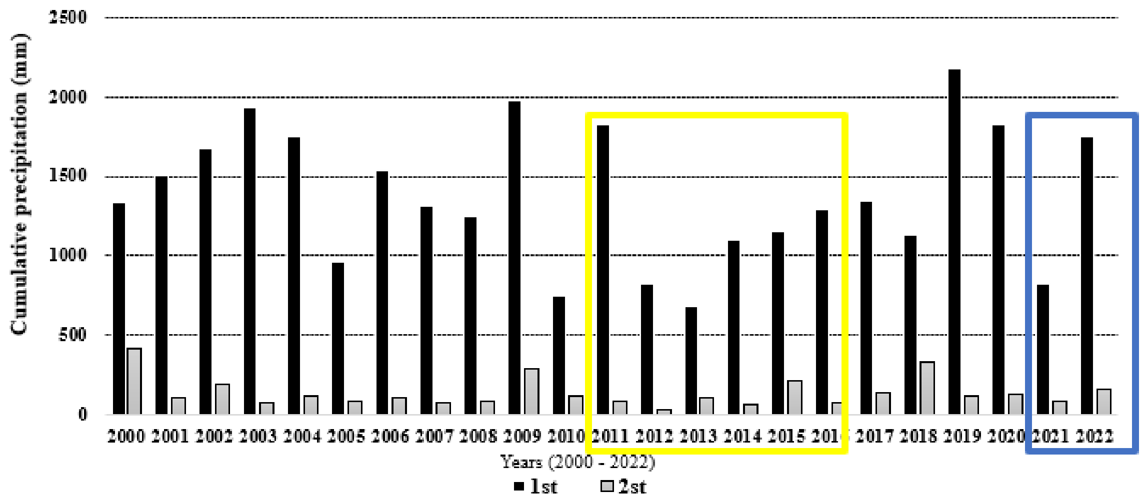


Figure 2 – Mean wind speeds (m/s) in the first (gray bar) and second (black bar) half of the years 2002 to 2022. Data obtained from INMET – Fortaleza station (2022).



The alternation between periods of high and low precipitation of the Pacoti river region in a times series (2000-2022) is shown in Figure 3. The precipitation rates were influenced mainly by changes in seasonal and interannual scales of climatic phenomena (DA SILVA *et al.*, 2013; COSTA; SILVA, 2017). The period from December 2011 to 2016 was considered a period of severe drought (Figure 3, yellow rectangles), intensified by the El Niño event in the year 2015, which generated a water crisis for the state (MARENGO; CUNHA; ALVES, 2016; BRITO *et al.*, 2017). In 2022, during the study year (Figure 3, blue rectangle), the mean precipitation in the first semester of the region was one of the highest values in relation to the previous year and to the period of severe drought (FUNCEME, 2022).

Figure 3 – Accumulated precipitation (mm) in Pacoti estuary region in the first (1st) and second (2st) semesters of the 2000 to 2022 years. The yellow rectangle highlights the period of severe drought in the region (2011 - 2016). The blue rectangle shows the sampling year (2022) and the previous year (2021). Data obtained from FUNCEME (2022).



The estuary of Pacoti river is in the metropolitan region of Fortaleza, capital of Ceará State (Northeast Brazil) and most of the river is included in an environmental protected area established in 2000-year (Decree N° 25.788/2000). The Pacoti River is barred along its course with three important dams, the Pacoti, Gavião and Riachão reservoirs. The construction of multiple dams was a solution for the semiarid region's physical characteristics and low availability of water resources for human consumption and economic activities (FRISCHKORN; ARAÚJO, SANTIAGO, 2003; LEMOS; OLIVEIRA, 2004). As 2022 was an atypical rainfall year compared to the historical drought series between 2011 and 2016, and the previous year (2021), the three dams had their water retention capacity completed and poured in the months of April, May, June, and July (Gavião) and May and June (Pacoti and Riachão) (FUNCEME, 2022) (Supplementary Material).

In the Pacoti estuarine systems, seagrass beds, sandy-muddy banks and mangrove roots provides habitats for mollusks, polychaetas, crustaceans and fishes (BARROS *et al.*, 2017; SILVA; FRANKLIN-JUNIOR; ROCHA-BARREIRA, 2017; GARCIA *et al.*, 2020; SOARES *et al.*, 2021). The near threatened longsnout seahorse (*Hippocampus reidi*) inhabit these estuarine waters as part of their life cycle (ROSA *et al.*, 2005; OSÓRIO; GODINHO; LOTUFO, 2011). Mangroves from this estuary are key ecosystems for artisanal fishers and shellfish gatherers who derive their income from the aquatic resources (LOPES *et al.*, 2021).

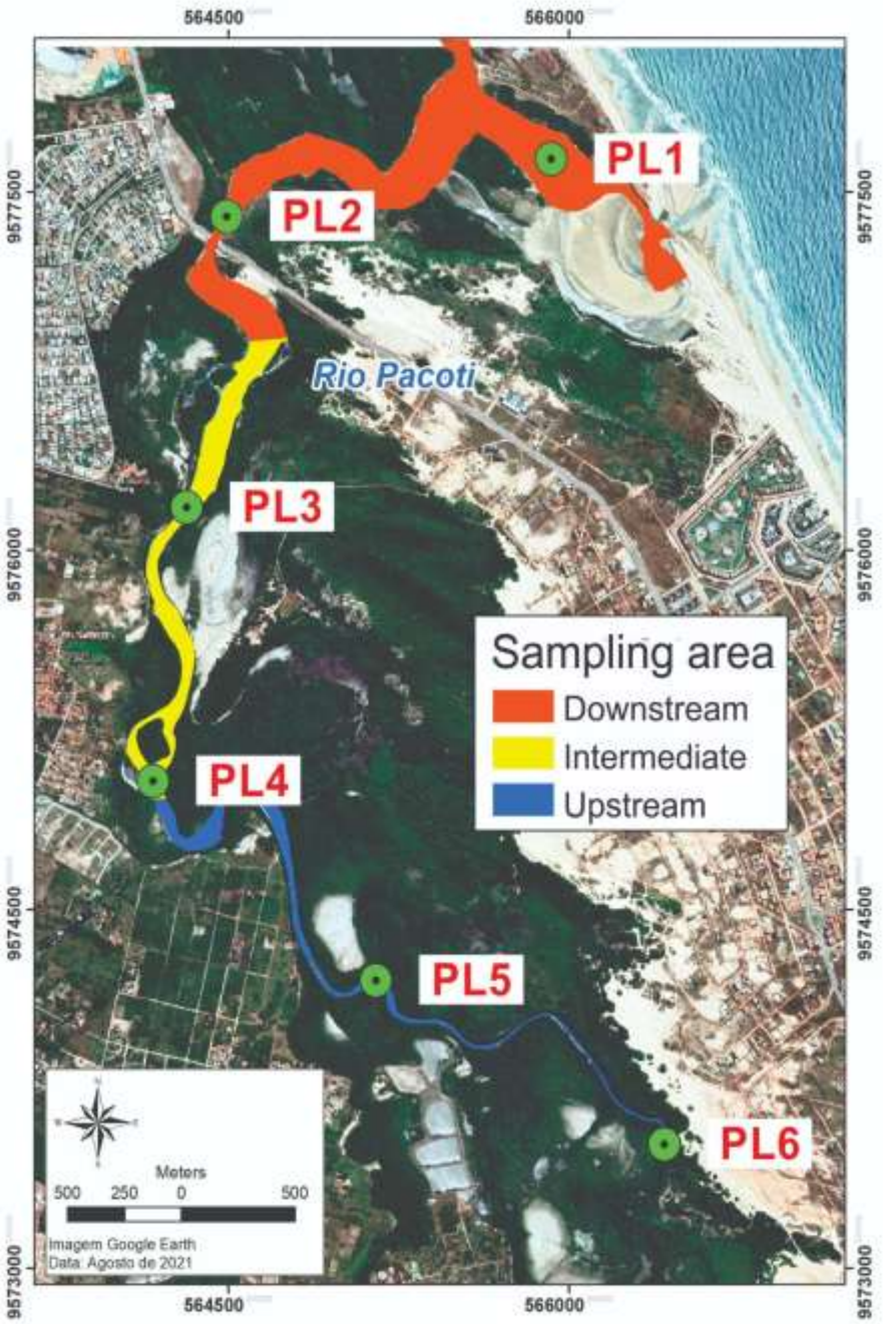
In the early 1960's, Pacoti mangroves were threatened by salt ponds that reduced mangrove area from 0.71 km² (1958) to 0.54 km² (1968), and then experienced an increase of mangrove area to 1.42 km², in 1999, and to 1.44 km², in 2004 (FERREIRA; LACERDA, 2016; LACERDA; MENEZES; MOLISANI, 2007). Nowadays, almost 20 years later, due to law protection, limit human activities and active restoration projects, Pacoti mangroves forests are increasing and expanding landward, offering ecosystem goods and services, such as blue carbon sequestration and aboveground and plant biomass storage (FERREIRA; ALENCAR; BEZERRA, 2019; FERREIRA; BEZERRA; MATHEWS-CASCON, 2019).

2.2 Field Sampling

Diurnal sampling of Ichthyoplankton was conducted through horizontal hauls using conical plankton nets (120 and 300 μm), at full moon of spring tide and during the ebb tide, in a single month of the rainy (April/2022), transition (August/2022) and dry (November/2022) seasons. The water volume filtered was determined by a flowmeter (General Oceanics®) attached to each net. Samples were fixed in Formol (4%) buffered with Borax. These hauls were conducted at six stations distributed along a salinity gradient (from downstream, near the river mouth, to upstream) (Figure 4).

These stations were grouped into three (3) different spatial zones, according to their distance from the river mouth: **Downstream**, close to the river mouth, comprising PL01 and PL02 stations; **Intermediate**, between the river mouth and upstream, comprising stations PL03 and PL04; **Upstream**, more influenced by freshwater, comprising stations PL05 and PL06 (Figure 4). This spatial zoning aim to test differences in spatial distribution patterns of fish eggs and larvae (The geographic coordinates of the sampling stations are available at the Supplementary material). Environmental data was measured during the sampling activities. At each station, values of salinity and temperature (°C) are obtained with a Castaway CTD (CastAway-CTD®), and Dissolved Oxygen (mg/L) are measured by means of a multiparameter probe (Hanna HI9829). In addition, water depth and transparency (m) are measured with a Secchi Disc.

Figure 4 – Ichthyoplankton sampling stations along the Tropical estuary of Pacoti river (Semiarid Coast of Brazil). The six stations are distributed along a salinity spatial gradient from the downstream (PL01 and PL02), passing through the intermediate zone (PL03 and PL04), to upstream (PL05 and PL06).



2.3 Laboratory procedures

Fish eggs and larvae were sorted, counted (number of organisms) and identified to the lowest possible taxonomic level according to the specialized literature (RÉ, 1999; RICHARDS, 2005; BONECKER *et al.*, 2014). Meristic and morphometric characteristics were used for the fish larvae taxonomic identification. Due to the morphological similarity of fish eggs, which makes it difficult to accurately identify the taxon, the identification was restricted to morphotypes, according to egg size (mm), presence or absence and amount of oil drops, and stage of development of the embryo (RÉ, 1999; RICHARDS, 2005; BONECKER *et al.*, 2014).

2.4 Data analysis

The density of eggs and larvae was calculated using ***D= N/V100m³***, where ***N*** = number of fish eggs or fish larvae, and ***V=a x n x c***, where ***a***=area of the net mouth, ***n***=number of rotations of the flowmeter and ***c***= calibration rate of the flowmeter. An average density was calculated from the two sampling points in each of the estuary zones.

Fish larvae was classified according to their developmental stage to elucidate the ways fish larvae use the estuarine system, based on MILLER; KENDALL JR (2009): **preflexion stage** (begins at the end of the yolk-sac larval stage and lasts until the notochord starts to flex), **flexion stage** (the tip of the notochord bends dorsally, and this stage ends when notochord flexion is complete) and **postflexion stage** (starts when notochord flexion is complete and ends when all of the fin rays have formed, and the juvenile stage begins). Fish larvae in very early developmental stages (yolk sac) or degraded were counted as unidentified taxa.

The larvae species was classified according to the ecological guilds proposed by POTTER *et al.* (2015): Marine (Marine straggler, Marine estuarine-opportunist, Marine estuarine-dependent); Estuarine (Solely estuarine, Estuarine and Marine, Estuarine and Freshwater and Estuarine migrant); Freshwater (Freshwater straggler, Freshwater estuarine-opportunist). The following references sources were used to identify the preferred habitats of fish adults in the Pacoti estuary and similar habitats (ANDRADE-TUBINO, RIBEIRO, VIANA, 2008; OSÓRIO, GODINHO, LOTUFO, 2011; GURGEL-LOURENÇO *et al.*, 2022).

3 RESULTS

3.1 Environmental variables

The tropical estuary of Pacoti river presented a typical salinity gradient of classic estuaries in 2022, with salinity values increasing from upstream to the downstream zone**,** during the rainy (U: 0.52 ± 0.1 → D: 34.5 ± 1.0), transition (U: 5.4 ± 0.0 → D: 35.3 ± 0.5) and dry (U: 13.0 ± 9.0 → D: 37.2 ± 0.0) seasons. Moreover, in the dry season, an increase in salinity values can be observed at the upstream zone (Rainy: 0.52 ± 0.1 → Transition: 5.4 ± 0.0 → Dry: 13.0 ± 9.0). A discrete increase in salinity in the downstream zone, during the dry season (37.2 ± 0.0) evidenced a value close to the salinity of the adjacent sea.

Regarding the water temperature in the Pacoti estuary, the values presented little variation (27 ± 0.2 to 30.2 ± 0.4) between the seasons and zones, which is a typical feature of tropical ecosystems. These values range between the downstream zone, during the transition season (27 ± 0.2 °C), and at the same zone, during the rainy season (30.2 ± 0.4 °C). The depth values obtained represent values typical of shallow estuaries, with values ranging between 0.8 ± 0.3 cm at the intermediate zone (rainy season) and 3.7 ± 0.7 cm at the downstream zone (rainy season).

The dissolved oxygen (DO) values showed a decreased gradient from the downstream to the upstream zone, during the rainy (D:6.1 ± 0.1 → I: 5.6 ± 1.3 → U: 4.9 ± 1.3) and during the dry season (D: 5.9 ± 0.2 → I: 3.8 ± 1.1 → U:3 4 ± 0.3). Interestingly, during the transition season, there was an increase in DO concentration from downstream to intermediate (6.1 ± 0.1 → 8.0 ± 0.6), but this value was reduced to 0 in the upstream zone. Water transparency showed a gradient of reduction from the clear waters of the downstream zone, near the river mouth, to the more turbid waters of the upstream zone. All these variables and their respective values are shown in Table 1.

Table 1 – Values of environmental variables (mean ± SE) recorded at Downstream, Intermediate and Upstream zones of the tropical estuary of Pacoti river (Semiarid Coast of Brazil). All these measurements were taken during the rainy (April/2022), transition (August/2022) and dry seasons (November/2022).

Rainy season			
Variables	**Downstream**	**Intermediate**	**Upstream**

Salinity	34.5 ± 1.0	9.2 ± 9.3	0.52 ± 0.1
Temperature (°C)	30.2 ± 0.4	29.1 ± 0.2	30.1 ± 0.4
Depth (m)	3.7 ± 0.7	0.8 ± 0.3	2.5 ± 1.1
Dissolved oxygen (mg/L)	6.1 ± 0.1	5.6 ± 1.3	4.9 ± 1.3
Water transparency (cm)	1.55 ± 0.4	0.3 ± 0.1	0.15 ± 0.1
Transition season			
Variables	**Downstream**	**Intermediate**	**Upstream**
Salinity	35.3 ± 0.5	19.8 ± 6.9	5.4 ± 0.0
Temperature (°C)	27 ± 0.2	27.8 ± 0.3	28.3 ± 0.2
Depth (m)	2.3 ± 0.6	1.1 ± 0.5	2.0 ± 0.9
Dissolved oxygen (mg/L)	6.1 ± 0.1	8.0 ± 0.6	0 ± 0
Water transparency (cm)	0.75 ± 0.1	0.4 ± 0.3	0.35 ± 0.1
Dry season			
Variables	**Downstream**	**Intermediate**	**Upstream**
Salinity	37.2 ± 0.0	34.6 ± 2.5	13.0 ± 9.0
Temperature (°C)	28.4 ± 0.1	28.8 ± 0.1	29.8 ± 0.3
Depth (m)	2.3 ± 1.1	0.9 ± 0.2	2.1 ± 1.0
Dissolved oxygen (mg/L)	5.9 ± 0.2	3.8 ± 1.1	3.4 ± 0.3
Water transparency (cm)	1.95 ± 0.5	0.5 ± 0.1	0.35 ± 0.1

3. 2 Fish eggs morphological characterization and density

A total of 71 eggs was sampled with 120 μm net and 353 eggs with the 300 μm net. The eggs were identified in five (5) morphotypes and Engraulidae, with the latter being easily identified due to the ellipsoid shape characteristic of the taxa. The illustrative representation of the morphotypes and eggs of the Engraulidae family, as well as the diagnostic features used are described in Table 2.

Table 2 - Morphotypes and diagnostic features of eggs sampled at three zones (Upstream, Intermediate and Downstream) in the tropical estuary of Pacoti river (Semiarid Coast of Brazil) during the rainy (April/2022), transition (August/2022) and dry (November/2022) seasons. Elaborated by Lucas Montenegro.

Eggs morphotypes		Diagnostic features
A		Ovoid shape with a diameter of 6 mm, absence of oil drop, smooth and transparent fecundation membrane, pigmented larvae in the dorsal region.
B		Ovoid shape with a diameter of 3.6 mm, presence of oil drop, larvae and yolk sac occupying almost the whole egg, absence of perivitelline space.
C		Ovoid shape, diameter 4.8 mm, opaque fecundation membrane, larvae and yolk sac occupy little space of the egg, perivitelline space 2.4 mm, absence of oil drop.
D		Ovoid shape with a diameter of 4.8 mm, smooth and transparent fertilization membrane, larvae and yolk sac occupies the center of the egg, perivitelline space of 1.2 mm.
E		Ovoid shape with diameter of 3.6 mm, embryo in the center of the egg at blastula stage, presence of oil drop and perivitelline space of 1.2 mm.

Engraulidae		Ellipsoid shape with 14.88 mm and 2.4 mm of perivitelline space. Larvae is long and attached to the yolk sac. Transparent membrane. Absence of oil drop.

Overall, the distribution of fish eggs was concentrated in the downstream zone near the river mouth, during all three seasons, with the 120 and 300 μm nets. However, during the transition season, fish eggs also occur at the intermediate zone in low densities, with both nets. During the rainy season, a total density of 7.05 eggs/100m^3 was found in the downstream zone with the 120 μm net, and 11.8 at this same zone with the 300 μm net. Among the egg morphotypes obtained, Morphotype E was the only one sampled with the 120 μm net during the rainy season, on the other hand, four morphotypes were sampled with the 300 μm net (A, B, C and D), with the Morphotype B and D (3.85 eggs/100m^3) presenting the highest densities.

During the transition season, we observe an increase in the density of eggs, in relation to the previous season (rainy). A total density of 67.35 eggs/100m^3 was found in the downstream zone (120 μm) and 0.85 eggs/100m^3 at the intermediate zone (120 μm), and 62.65 eggs/100m^3 at the downstream zone (300 μm) and 4.4 eggs/100m^3 at the Intermediate zone (300 μm). Morphotype E and Engraulidae were sampled with the120 μm net, and Morphotype A, Morphotype E and Engraulidae were sampled with the 300 μm net. The highest density values during this season were obtained by Morphotype E with the 120 (65 eggs/100m^3) and 300 μm net (58.7 eggs/100m^3). Engraulidae eggs only appeared during this season.

The distribution of eggs in the dry season showed a similar pattern to the rainy period, being concentrated only in the downstream zone. We also observed a decreased in the density of eggs, in relation to the previous season (transition). A total of 17.85 eggs/100m^3 was found in the downstream zone with the 120 μm net, and a total of 10.8 eggs/100m^3 was found in this same zone with the 300 μm net. Among the eggs morphotypes obtained in Pacoti estuary, this season presented the occurrence of Morphotype A and Morphotype E with both nets. The highest density values during this season were obtained by Morphotype E with the 120 (14.65 eggs/100m^3) and 300 μm net (6.5 eggs/100m^3). Therefore, among the egg morphotypes, Morphotype E was the most frequent throughout the year with the 120 mesh net and Morphotype A and E were more frequent throughout the year with the 300 mesh net. The density values (eggs/100m^3), the morphotypes identified, as well as the distribution of these organisms along the estuarine zones are presented in Table 3.

Table 3 – Mean density (N/100m³) of fish eggs at the three zones (Downstream, Intermediate and Upstream) of the tropical estuary of Pacoti river (Semiarid Coast of Brazil), during the rainy, transition and dry seasons. The densities are also shown in relation to both mesh nets (120 and 300 μm).

	Morphotype	Downstream	Intermediate	Upstream	Morphotype	Downstream	Intermediate	Upstream
Rainy season	A	0	0	0	A	1.9	0	0
	B	0	0	0	B	3.85	0	0
	C	0	0	0	C	2.2	0	0
	D	0	0	0	D	3.85	0	0
	E	7.05	0	0	E	0	0	0
	Engraulidae	0	0	0	Engraulidae	0	0	0
	TOTAL	**7.05**	**0**	**0**	**TOTAL**	**11.8**	**0**	**0**
	Pattern of distribution	**D > I and U**			**Pattern of distribution**	**D > I and U**		
Transition season	A	0	0	0	A	2,05	0	0
	B	0	0	0	B	0	0	0
	C	0	0	0	C	0	0	0
	D	0	0	0	D	0	0	0
	E	65	0	0	E	58,7	3,25	0
	Engraulidae	2.35	0.85	0	Engraulidae	1,9	1,15	0
	TOTAL	**67.35**	**0.85**	**0**	**TOTAL**	**62.65**	**4.4**	**0**
	Pattern of distribution	**D > I > U**			**Pattern of distribution**	**D > I > U**		
Dry season	A	3.2	0	0	**A**	4.3	0	0
	B	0	0	0	**B**	0	0	0
	C	0	0	0	**C**	0	0	0
	D	0	0	0	**D**	0	0	0
	E	14.65	0	0	**E**	6.5	0	0
	Engraulidae	0	0	0	**Engraulidae**	0	0	0

	TOTAL	**17.85**	**0**	**0**	**TOTAL**	**10.8**	**0**	**0**
	Pattern of distribution	**D > I and U**			**Pattern of distribution**	**D > I and U**		

3.3 Fish larvae density and taxonomic composition

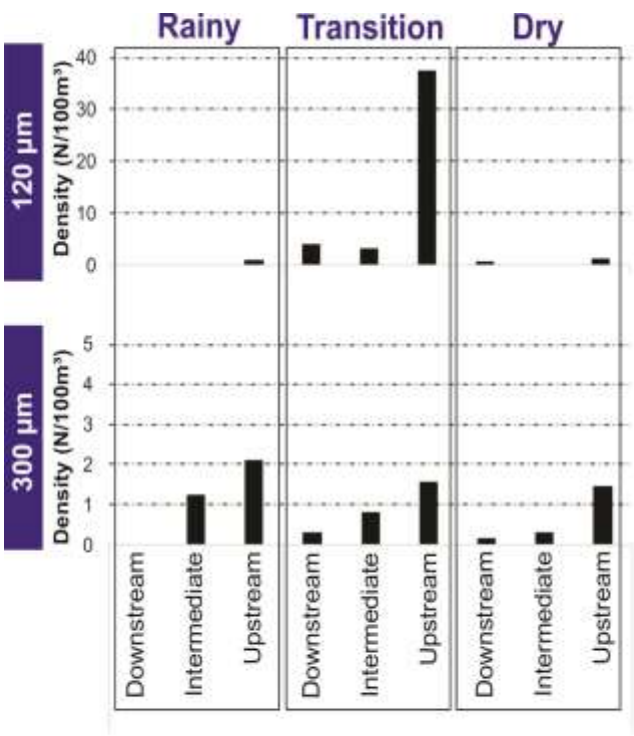
A total of 27 fish larvae were sampled with 120 μm net and 24 fish larvae with the 300 μm net. In contrast to what was observed for eggs, the highest densities of fish larvae were mainly found at the intermediate and upstream zones, especially the latter. Among the 51 fish larvae sampled, it was found 6 unidentified larvae.

The rainy season presented low densities of fish larvae. During this season, fish larvae were recorded only at upstream zone (0.8 larvae/100m³) with the 120 μm net, and at intermediate (1.25 larvae/100m³) and upstream (2.1 larvae/100m³) zones with the 300 μm net. Therefore, fish larvae were mainly found in the upstream zone with both nets.

The transition season presented an increase in the density of fish larvae, in comparison with the previous season (rainy). During the transition season, fish larvae were recorded in all three zones, but with different density values. In this season, the highest density values were obtained at the upstream zone (37.65 larvae/100m³ and 1.55 larvae/100m³) with the 120 and 300 μm nets, respectively. Thus, just like the previous season, the upstream area was the one where the fish larvae were concentrated.

The dry season presented a low density of fish larvae, in comparison with the previous season and likewise the dry season. Fish larvae were also distributed in the three zones, during the dry season, however, this pattern only occurred with the results obtained with the 300 μm net. Also, the highest densities values were obtained at the upstream zone (1.05 larvae/100m³ and 1.45 larvae/100m³) with both 120 and 300 μm, respectively. Regarding the results obtained from the 120 μm mesh net, during the dry season, fish larvae were found only at the downstream (0.65 larvae/100m³) and upstream zones (1.05 larvae/100m³). Despite the low density of fish larvae, the upstream zone also showed high densities, likewise the other seasons, and compared to the other zones in the same season. The graphical representation of these results is shown in Figure 5.

Figure 5 – Mean density (N/100m³) of fish larvae at the three zones (Downstream, Intermediate and Upstream) of the tropical estuary of Pacoti river (Semiarid Coast of Brazil), during the rainy, transition and dry seasons. The densities are shown in relation to both mesh nets (120 and 300 μm).



The assemblage of fish larvae in the tropical estuary was composed by eight (8) families distributed in 8 taxa, which includes four (4) species, three (3) genera and one (1) family. All these species are classified as ''Least Concern - LC'' based on the Red List Assessment (IUCN). According to the characteristics of the adults, these taxa are classified into the following ecological guilds: Solely estuarine or Resident (*Atherinella brasiliensis*, *Gobionellus oceanicus* and *Sphoeroides testudineus*), Marine estuarine-opportunist (*Strongylura* sp. and *Hyporhamphus unifasciatus*) and Marine Estuarine-dependent (*Eucinostomus* sp., Sciaenidae sp1 and *Anchovia clupeoides*) (Table 4).

Table 4 – Composition (Family and species), Common name, red list assessment (IUCN) and Ecological guilds of the fish larvae assemblage of the Tropical estuary of Pacoti river (Semiarid Coast of Brazil) during the rainy, transition and dry seasons, with 120 and 300 µm mesh nets.

Family	Species	Common name	Red List assessment (IUCN)	Ecological guilds
Atherinopsidae	*Atherinella brasiliensis*	Robust silverside	Least concerned (LC)	Solely estuarine (Resident)
Belonidae	*Strongylura* sp.	Needlefish	Least concerned (LC)	Marine estuarine-opportunist
Engraulidae	*Anchovia clupeoides*	Zabaleta Anchovy	Least concerned (LC)	Marine Estuarine-dependent
Gerreidae	*Eucinostomus* sp.	Mojarra	Least concerned (LC)	Marine Estuarine-dependent
Gobiidae	*Gobionellus oceanicus*	Highfin Goby	Least concerned (LC)	Solely estuarine (Resident)
Hemiramphidae	*Hyporhamphus unifasciatus*	Atlantic Silverstripe Halfbeak	Least concerned (LC)	Marine estuarine-opportunist
Sciaenidae	Sciaenidae sp1	Croakers	-	Marine Estuarine-dependent
Tetraodontidae	*Sphoeroides testudineus.*	Pufferfish	Least concerned (LC)	Solely estuarine (Resident)

The rainy season presented the occurrence of *A. brasiliensis* (0.45 larvae/100m^3) and *Eucinostomus sp.* (0.35 larvae/100m^3) at the upstream zone with the 120 µm net. Regarding the 300 µm net, *A. clupeoides* (0.5 larvae/100m^3) was found at the intermediate zone and *Eucinostomus sp*. (1.75 larvae/100m^3) at the upstream zone. On the other hand, the transition season presented the occurrence of more species compared to the rainy season and in all three zones.

During the transition season, the species distribution with the 120 μm net was according to the following pattern: the upstream zone presented the occurrence of *A. clupeoides* (0.65 larvae/100m³), *A. brasiliensis* (19.95 larvae/100m³), *G. oceanicus* (12.7 larvae/100m³), *Strongylura sp.* (2.5 larvae/100m³) and *H. unifasciatus* (2.5 larvae/100m³). Moving on to the intermediate zone, this region presented the occurrence of *A. brasiliensis* (0.85 larvae/100m³) and *H. unifasciatus* (0.65 larvae/100m³). The downstream zone presented the occurrence of *A. brasiliensis* (2.75 larvae/100m³) and *A. clupeoides* (1.2 larvae/100m³).

During the dry season, there was a reduction in the number of species compared to the previous season. This season presented the occurrence of *A. clupeoides* (0.65 larvae/100m³) at the downstream zone and *Eucinostomus sp.* (0.7 larvae/100m³) and *A. brasiliensis* (0.35 larvae/100m³), with the 120 μm net. Regarding the 300 μm net, *A. clupeoides* was found at the upstream (0.6 larvae/100m³) and intermediate (0.1 larvae/100m³) zones, *Eucinostomus* sp. was found at the Upstream zone (0.35 larvae/100m³), *A. brasiliensis* was found at the upstream (0.55 larvae/100m³) and intermediate (0.2 larvae/100m³) zones, Sciaenidae sp1 was only found only at the intermediate zone (0.15 larvae/100m³). The species distribution along the zones in the tropical estuary of Pacoti river and in relation to the mesh sizes (120 and 300) are presented in Table 5.

Table 5 – Mean density (N/100m³) of fish larvae species along the three zones (Downstream, Intermediate and Upstream) of the tropical estuary of Pacoti river (Semiarid Coast of Brazil), during the rainy, transition and dry seasons. The densities are also shown in relation to both mesh nets (120 and 300 µm).

		Rainy			Transition			Dry		
120µm	**Taxa**	**Downstream**	**Intermediate**	**Upstream**	**Downstream**	**Intermediate**	**Upstream**	**Downstream**	**Intermediate**	**Upstream**
	Anchovia cluepeoides	0	0	0	1.2	0	0,65	0,65	0	0
	Eucinostomus sp.	0	0	0.35	0	0	0	0	0	0.7
	Atherinella brasiliensis	0	0	0.45	2.75	0.85	19.95	0	0	0.35
	Gobionellus oceanicus	0	0	0	0	0	12.7	0	0	0
	Strongylura sp.	0	0	0	0	0	25	0	0	0
	Hyporhamphus unifasciatus	0	0	0	0	0.65	2.5	0	0	0
	Unidentified	0	0	0	0	0.85	0	0	0	0
	TOTAL	**0**	**0**	**0.8**	**3.95**	**2.35**	**38.3**	**0.65**	**0**	**1.05**
	Pattern of distribution	**D and I > U**			**D > I < U**			**D < I > U**		
300µm		**Rainy**			**Transition**			**Dry**		
	Taxa	**Downstream**	**Intermediate**	**Upstream**	**Downstream**	**Intermediate**	**Upstream**	**Downstream**	**Intermediate**	**Upstream**
	Anchovia cluepeoides	0	0.5	0	0.3	0.8	0	0	0.1	0.6

	Eucinostomus sp.	0	0	1.75	0	0	0	0	0	0.35
	Atherinella brasiliensis	0	0	0	0	0	0.5	0	0.2	0.55
	Gobionellus oceanicus	0	0	0	0	0	0,6	0	0	0
	Sphoeroides testudineus	0	0	0	0	0	0,5	0	0	0
	Sciaenidae sp1	0	0	0	0	0	0	0	0,15	0
	Unidentified	0	0.75	0.35	0	0	0	0,15	0	0
	TOTAL	**0**	**1.25**	**2.1**	**0.3**	**0.8**	**1.6**	**0.15**	**0.45**	**1.5**
	Pattern of distribution	**D < I < U**			**D < I < U**			**D < I < U**		

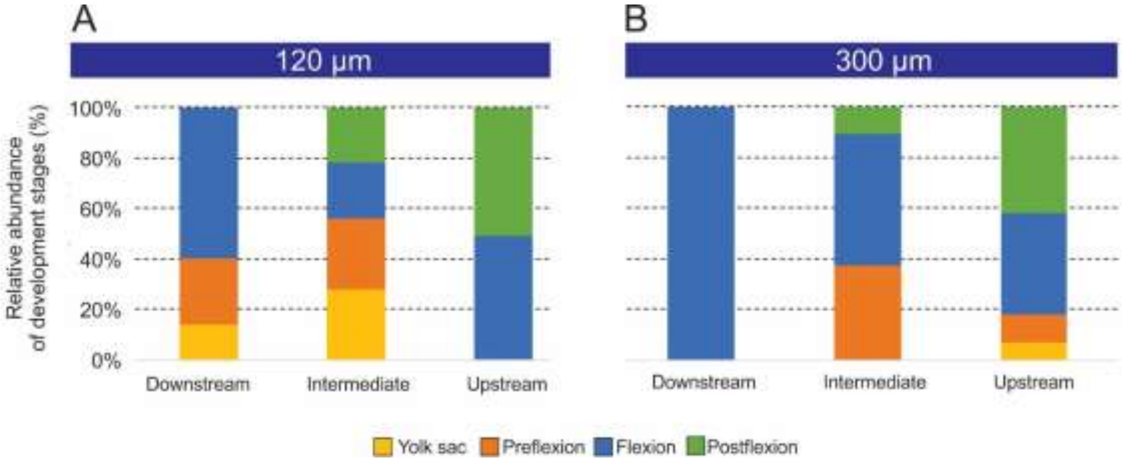
3.4 Stages of development and Ecological guilds

The sampling in the Pacoti estuary showed a total of 15 larvae in postflexion, 21 larvae in flexion, 8 larvae in preflexion, and 3 larvae in the yolk sac stage, evidencing the dominance of larvae in more advanced stages of development.

Abundant fish larvae in flexion (49%) and postflexion (51%) stages were concentrated upstream, while fish larvae in all stages of development, Yolk sac (28%), preflexion (28%), flexion (22%) and postflexion (22%), were found in the intermediate zone. Fish larvae in downstream zone, near the river mouth, was found in yolk sac (14%), preflexion (26%) and flexion stages (60%). Therefore, a pattern of yolk sac, preflexion and flexion stages were found in the downstream and intermediate zones, where the density of fish larvae were the lowest, and postflexion and flexion larvae were mainly found upstream, where the highest density values were obtained. These results were obtained with the 120 µm net.

Conversely, with the 300 µm net, all the stages of development, yolk sac (7%), preflexion (11%), flexion (40%) and postflexion (42%), were found Upstream, where the highest densities of fish larvae were obtained. Preflexion (37%), flexion (52%) and postflexion (11%) larvae were found in the intermediate zone, but in the downstream zone, fish larvae were in flexion stage (100%). Therefore, with the 300 µm mesh net, we found a pattern of fish larvae in flexion stage distributed throughout the estuarine system and all developmental stages upstream, where high larvae density values were found. The data of relative abundance (%) of the development stages in the sampling areas and in both nets are represented in Figure 6.

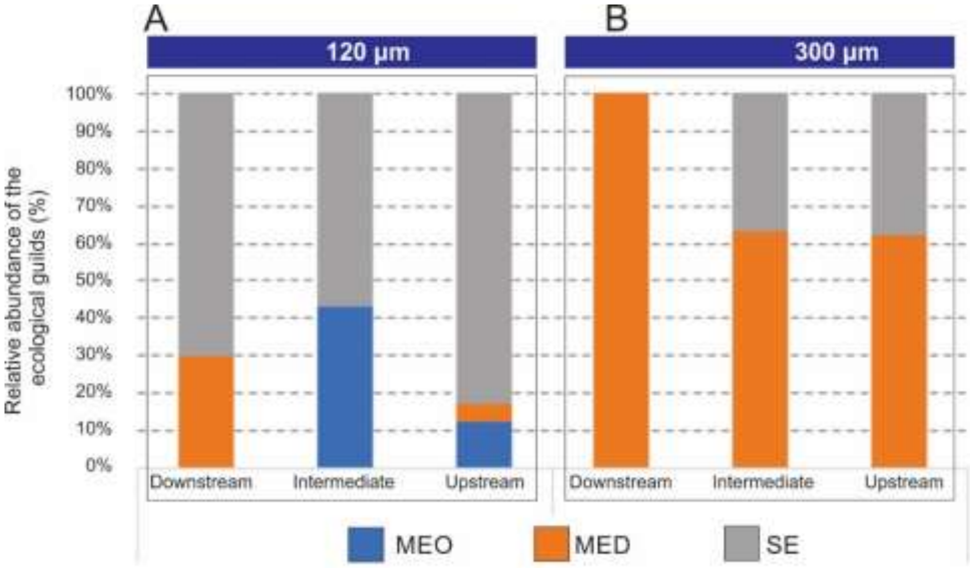
Figure 6 – Relative abundance (%) of Stages of development from the Tropical estuary of Pacoti river (Semiarid Coast of Brazil) at the three zones (downstream, intermediate, and upstream), with both 120 µm and 300 µm net.



The fish larvae species at the Pacoti estuary (Semiarid Coast of Brazil) were only classified as guilds that belong to the marine and estuarine environments. The absence of freshwater species, even though the sampling year was influenced by high precipitation and freshwater discharge, is remarkable for this semiarid estuarine system. Both 120 and 300 µm nets sampled different number of species, however solely estuarine (resident) species were found with both nets (Figure 7).

Marine estuarine-opportunist species were found in upstream (12%) and intermediate (43%) zones, whilst Marine estuarine-dependent species were found at downstream (30%) and upstream (5%) zones. Solely estuarine species were found in all zones of the Pacoti estuarine systems, downstream (70%), intermediate (57%) and upstream (83%). This was the pattern found with the 120 µm mesh size. Conversely, only Marine estuarine – dependent and solely estuarine species were sampled with the 300 µm mesh size, and pattern of absence of freshwater species was also found. Marine estuarine – dependent species were found distributed along the estuarine system: downstream (100%), intermediate (63%) and upstream (62%). Solely estuarine species were found in intermediate (37%) and upstream (38%). The data of relative abundance (%) of the development stages in the sampling areas and in both nets are represented in Figure 7.

Figure 7 – Relative abundance (%) of Ecological guilds from the Tropical estuary of Pacoti river (Semiarid Coast of Brazil) at the three zones (downstream, intermediate, and upstream), with both 120 µm and 300 µm net. MEO: Marine estuarine-opportunist, MED: Marine estuarine-dependent, SE: Solely estuarine. Freshwater taxa were absent.



4 DISCUSSION

The results of this baseline highlight the spatial and seasonal distribution of fish eggs and larvae along a shallow and short tropical estuary influenced by the semiarid climate, during a year of heavy precipitation. Overall, even though mangroves are known as nursery and feeding zones by early life stages of fish (DIAS *et al.*, 2022; TARIMO *et al.*, 2022), during this year, the analyzed mangrove estuarine ecosystem presented a low number of taxa (N = 8 taxa of larvae and 6 taxa of eggs) and the density of ichthyoplankton was also low for both mesh sizes of net (120 and 300 µm).

4.1. Environmental factors and spatial distribution patterns along the estuary

Fish eggs were concentrated in high densities in the downstream zone, near the river mouth, but was also found in low densities, in the intermediate zone, during the transition season. This pattern for fish eggs in Pacoti estuary can be explained by the interaction of three factors: I - the proximity of this zone to the coastal waters, where marine fishes spawn, II - the mixing of marine/estuarine waters to replenish dissolved oxygen (DO), which is essential for egg development, and III - the flushing of fish eggs out of intermediate and upstream zones, mainly during events of heavy precipitation and freshwater release by dams.

The transport of organisms between the estuarine and marine environments through tidal variation (import/export dynamics) is well known for several estuarine organisms, such as crabs, shrimps, and fish (CAMPOS, GARCIA, FORTALEZA, 2016; MACURA *et al.*, 2019; LEWIS *et al.*, 2020; NÓBREGA *et al.*, 2021). This physical process can explain the presence of fish eggs in the downstream zone of the estuary, since this dynamic changes daily. Concerning ichthyoplankton, this change can impact the diversity, distribution, and density, mainly because it affects the survival of these organisms, since they can be removed to unsuitable areas (BARLETTA-BERGAN; BARLETTA, SAINT-PAUL, 2002a).

In 2022, during the rainy, Pacoti dams (Riachão, Gavião and Pacoti) poured and released freshwater into the system, but these events extended until close to the transition season, when precipitation was reduced. The water volume in the reservoirs of Ceará, such as those that compose the Pacoti system, reached their maximum capacity and spilled during the first half of the year (FUNCEME, 2022). The results of this extreme precipitation event may have limited the intrusion of salt wedge into the system.

Despite being highly impacted by human activities, coastal zones are known to be spawning zones to marine fishes that depend on the ecological connectivity between the mangroves and the coast to complete their life cycle (HUTCHINGS *et al.*, 2002). There are some examples of this process and implications for the survival of the species, since spawning in productive environments prevents newly hatched larvae from being transported to other areas, where they cannot find food when the exogenous feeding phase begins (OOI; CHONG, 2011; VANALDERWEIRELDT *et al.*, 2020). Although only genetic analysis can confirm the species related to Morphotype E, taxon with the highest density eggs, the occurrence during all seasons in downstream zone implies that this taxon has a reproductive strategy of spawning close to the coast.

It is important to emphasize that high concentrations of dissolved oxygen (DO) were found in the downstream zone during the rainy (6.1 ± 0.1 mg/L) and dry (5.9 ± 0.2 mg/L) seasons, and in the intermediate zone (8.0 ± 0.6 mg/L), during the transition season. The mixing of the oligotrophic water from the adjacent sea with the estuarine water through tidal variation, not only brings fish eggs from the adjacent sea, but also replenishes the DO (BOLLE *et al.*, 2009).

Although fish larvae were found along all the sampling zones, the upstream zone presented the highest densities values throughout the year, with both 120 and 300 μm nets. This zone was characterized by low levels of oxygen, salinity, and water transparency. This pattern can mainly be attributed to feeding behavior and the stages of development where these larvae were found.

Mangroves are productive ecosystem in tropical estuaries used by early life stages of fish as a feeding zone, where they feed on zooplankton and phytoplankton prey and acquire energy to grow and develop (LIMA; BARLETTA, 2016; WHITFIELD, 2017). Initially, feeding is endogenous, through maternal resources concentrated in the yolk sac, which is gradually consumed, while the larvae move on to exogenous feeding (YÚFERA; DARIAS, 2007). With the aid of complete fins (postflexion stages), fish larvae can actively search for food in more turbid waters when they acquire energy to recruit (DAVID – RUALES; FRACALOSSI; TORRES, 2018).

Fish larvae in the upstream zone of Pacoti estuary, where high densities of larvae were found, are mostly in flexion and postflexion stages. Based on the match-mismatch hypothesis, the upstream turbid zone of Pacoti estuary may serve as feeding area for fish larvae, where they take advantage of zooplankton biomass and organic particles (CUSHING, 1990; FARIA; CHICHARO, 2006; OOI; CHONG, 2011). Furthermore, this hypothesis may also be associated with the higher number of species during the transition season and the high densities of fish eggs and larvae, as the end of the rainy season may stabilize nutrient inputs and allow for consumption by breeding adults and larvae.

4.2. Fish larval species and ecological features

Fish larvae assemblage of Pacoti estuarine system was composed of species found only in estuaries (*Atherinella brasiliensis*, *Gobionellus oceanicus* and *Sphoeroides testudineus*), species that regularly enter estuaries in substantial numbers, but use to varying degrees coastal marine waters as alternative nursery areas (*Strongylura sp.* and *H. unifasciatus*), and species that requires sheltered estuarine habitats (*Eucinostomus sp*., Sciaenidae sp1 and *A. clupeoides*) (POTTER *et al.*, 2015). Nutrient input into the estuarine system during heavy precipitation events benefit resident species and attract fish larvae from marine environments that use visual and olfactory cues to seek out these productive areas, as the marine and estuarine resident species found in this study (WHITFIELD, 2005; SANTOS; RAMOS; BONECKER, 2017a).

The Zabaleta anchovy (*Anchovia clupeoides*) are abundant component of ichthyoplankton assemblages in mangroves and the continental shelf of the Semiarid Coast of Brazil (BARLETTA-BERGAN; BARLETTA; SAINT-PAUL, 2002b; MOTA *et al*., 2017). These species feed on plankton and can tolerate a wide range of salinities, therefore, can be found from hypersaline environments to brackish water and, occasionally, freshwater environments (CARPENTER, 2002). The Engraulidae family in which this species is included is characterized by continuous spawning events (iteroparity) along the coast and they use estuaries during all or part of their life cycle for growing and development, which is why this species occurred in Pacoti estuary throughout the year (OSÓRIO; ACERO, 1996; LIMA; BARLETA, 2016).

Eucinostomus sp. is a coastal marine genus of Mojarras frequently found as adults in the mangrove roots of Pacoti estuary, where they take advantage of the productivity of these systems also during early stages of development (OSÓRIO; GODINHO; LOTUFO, 2011). In this study, larvae of this species were found only in the upstream zone, during the rainy and dry seasons. However, this species was also found in a hypersaline estuary (Max. of 53.2 ± 8.38) of the Semiarid Coast of Brazil, during the dry season, which shows a wide tolerance of this taxa to salinity variation (FRANCO *et al.*, 2012; ARRUDA JÚNIOR *et al*., 2023).

The Brazilian silverside (*Atherinella brasiliensis*) is a generalist and opportunistic predator and estuarine resident species found in high relative density in the Pacoti estuary during all seasons (CONTENTE; STEFANONI; SPACH, 2011). Early life stages of this species prey on zooplankton and is possibly constituent of a great relevance in the estuarine trophic web (DA ROCHA; SILVA-FALCÃO; SEVERI, 2008; BRITO *et al.*, 2019). Furthermore, this species can be found at different salinity levels (euryhaline), between classic and hypersaline estuaries, since it can alter its diet to osmoregulation more efficiently and complete its life cycle (SALES *et al.*, 2018; VIEIRA JÚNIOR *et al.*, 2021).

Gobies, such as the highfin goby (*Gobionellus oceanicus*), are abundant components of the ichthyofauna in tropical estuaries (GOMES; BONECKER, 2014). Larvae of this species was found only during the transition season in the upstream zone of the Pacoti estuary, when oxygen level was 0. This is an estuarine specie that can withstand hypoxia and survive freshwater environments (ÁLVARO; HERRERA; ANGULO, 2018; ARAÚJO; AGUILAR; DA CRUZ, 2021). They can be found in postflexion stages far from the river mouth of tropical estuaries, likewise we found in this study, searching for food to develop and grow (GOMES; CAMPOS; BONECKER, 2014).

Sphoeroides testudineus is a common estuarine species of the Brazilian coast, known as Pufferfishes, with records of adults in the mangroves of the tropical estuary of Pacoti (EKAU *et al.*, 2001; NAMIKI *et al.*, 2007; FÁVARO *et al.*, 2009; OSÓRIO; GODINHO; LOTUFO, 2011). Despite the occurrence of this species during the transition season (300 µm), the absence of studies on the reproductive biology of pufferfish in the Pacoti estuary makes it difficult to understand this outcome. However, research on the presence of endocrine disruption chemicals (EDC) in the sediments of Pacoti and the confirmation that these contaminants negatively affect the reproductive system of this species may explain the absence of larvae and, in addition, draws attention to xenobiotic pollution, especially during a year of high freshwater input from the continent (PIMENTEL *et al.*, 2016).

Needlefish larvae (*Strongylura* sp.) were found during the transition season in the Pacoti estuary, where two species of this genus are found: *S. marina* (Walbaum, 1792) and *S. timucu* (Walbaum, 1792) (GURGEL-LOURENÇO *et al*., 2022). Most of the species of this genus are estuarine-marine and widely spread in tropical and subtropical waters, but there are also freshwater species (COLETTE *et al.*, 2018). Another species that is also included in the order Beloniformes is the Atlantic Silverstripe Halfbeak (*H. unifasciatus*) which was found in the intermediate and upstream zones of the Pacoti estuary, during the transition season. This species is widely distributed in coastal areas of the America where they feed on zooplankton and phytoplankton (GUERRA *et al.*, 2021; CASTILLO-RIVERA; MORGADO-DUEÑAS, 2022) and has nutritional and economic value (GUERRA *et al.*, 2022).

The lowest possible taxonomic level for the identification of the Sciaenidae larvae found was family since the organism was in a very early stage of development (preflexion). This taxon was only found in the downstream zone of Pacoti estuary, during the dry season (300 µm). Croakers (Sciaenidae) are an important resource for small-scale artisanal fisheries in Brazil, but the risk of extinction due to fishing pressure is real (CHAO *et al.*, 2015). Fish larvae of this family can be found in freshwater, estuarine and marine environments, where they look for more sheltered habitats to develop and grow (COSTA *et al.*, 2012; CHAVES *et al.*, 2019; COSTA *et al*., 2020).

Larvae of freshwater species were absent in Pacoti estuary during all seasons. Despite the occurrence of freshwater fish larvae during rainy periods, periods of high freshwater input, or in estuaries dominated by large rivers (ARÉVALO-FRÍAS; MENDOZA - CARRANZA, 2011; CORREA - HERRERA *et al.*, 2017; ROUSSEAU; BLANCHARD; GARDEL, 2017), some factors can explain the absence of this organisms in some estuaries.

Osmotic stress due to fluctuating salinity values within the estuary hinders the ability to compete for food and survive in the estuarine environment, especially during the dry season (WHITFIELD, 2015; SLOTERDIJK *et al.* 2017). Furthermore, freshwater species commonly spawn in upstream areas, where eggs and larvae are retained in aquatic macrophytes to complete development in calmer and less saline waters (MONTOYA-MAYA; STRYDOM, 2009). Therefore, the absence of freshwater fish larvae in Pacoti estuary may be due to reproductive strategies, such as the use of spawning and developmental areas far from the upstream zone, where sampling was not possible due to navigational limitations (8 km from the mouth to the last sampling station).

This is the first intra-annual survey conducted in the tropical estuary of Pacoti (an Environmental Protection Area), focusing on understanding the dynamics of the ichthyoplankton community. In fact, previous studies about other plankton organisms in this estuary revealed the influence of freshwater discharge and salinity as structuring factors of this community (BARROSO; BECKER; MELO, 2016; GARCIA *et al.*, 2020) like other tropical estuaries (ROUSSEAU; BLANCHARD, GARDEL, 2017; SANTOS; SEVERI, 2019; ARA *et al.*, 2020). Studies conducted in the Semiarid Coast of Brazil have also revealed low densities and diversity of fish eggs and larvae in coastal ecosystems of this region, which is a feature of these communities in this region (MOTA *et al.*, 2014; MOTA *et al*., 2017; COSTA *et al*., 2020; ARRUDA JÚNIOR *et al*., 2023).

4 CONCLUSION AND FINAL REMARKS

Our baseline revealed the intra-annual influence of different precipitation regimes (rainy, transition and dry) in the estuarine distribution and composition of fish eggs and larvae of the tropical estuary of Pacoti river. This is the first assessment of ichthyoplankton in a particular and poorly studied region of Brazil, the Semiarid Coast. The ichthyoplankton assemblage is composed of estuarine resident and marine species, which reinforces the use of the estuary as an important nursery area for these organisms.

Extreme climatic events (heavy precipitation or severe droughts) are expected for the region studied, which can negatively compromise the nursery function of these systems and affect fish stocks. In this sense, baseline studies like this are important as a basis for future monitoring and long-term analysis of the influence of these extreme events on the recruitment variability of fish and to mitigate the impacts of climate change.

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Supplementary Material

Table 6 – Dams capacity (hm³) built along the Pacoti River, mean volume (hm³) of water occupying the reservoir in each month of 2022, and relative percentage (%) occupied by water. Data obtained from the Hydrological Portal of Ceará (FUNCEME - 2022).

	Gavião reservoir			Pacoti reservoir			Riachão reservoir		
Month (2022)	**Capacity (hm³)**	**Mean Vol. (hm³)**	**Vol. perc. (%)**	**Capacity (hm³)**	**Mean Vol. (hm³)**	**Vol. perc. (%)**	**Capacity (hm³)**	**Mean Vol. (hm³)**	**Vol. perc. (%)**
Jan.	33.3	28.3	85%	380	86.0	23%	47.92	13.9	29%
Feb.	33.3	28,6	86%	380	89.5	24%	47.92	13.6	28%
Mar	33.3	29.9	90%	380	127.8	34%	47.92	16.7	35%
Apr.	33.3	30.1	90%	380	264.0	69%	47.92	35.2	73%
May.	33.3	33.5	100%	380	362.2	95%	47.92	48.3	100%
Jun.	33.3	33.5	100%	380	371.9	98%	47.92	49.8	100%
Jul.	33.3	33.4	100%	380	367.2	97%	47.92	49.1	100%
Aug.	33.3	32.8	99%	380	348.4	92%	47.92	46.5	97%
Sep.	33.3	30.4	91%	380	321,4	85%	47.92	321.4	85%
Oct.	33.3	28.7	86%	380	306.8	81%	47.92	306.8	81%
Nov.	33.3	28.7	86%	380	263.2	69%	47.92	263.2	69%
Dec.	33.3	28.8	87%	380	233.7	62%	47.92	233.7	62%

Table 7 – Geographic coordinates of the sampling stations in the Tropical estuary of Pacoti river, grouped in three zones.

		Coordinates (WGS 84 Zona 24 Sul - UTM)	
	Sampling stations	**Latitude**	**Longitude**
Downstream	**PL01**	565910	9577637
	PL02	564504	9577363
Intermediate	**PL03**	564401	9576301
	PL04	564153	9575055
Upstream	**PL05**	565174	9574189
	PL06	566427	9573442

GENERAL CONCLUSIONS AND FINAL CONSIDERATIONS

The results of this research presented important conclusions for the study of ichthyoplankton in tropical estuaries. The three chapters was based on a general analysis (primary data) and compilation of studies on fish eggs and larvae in these coastal environments. Moreover, they served as a direction for the study of these organisms in a region that is unique and particular in terms of biodiversity and ecological processes in the Tropical South Atlantic, the Semiarid Coast of Brazil.

In Chapter 01, we demonstrated that tropical estuaries are key coastal ecosystems for the development, survival, and recruitment of early life stages of freshwater, estuarine, and marine fishes. Studies on this topic, however, are still few compared to estuaries in temperate regions. Furthermore, lack of standardization in sampling and presentation of results is still a problem when it is necessary to compare studies of estuarine systems between regions. In this sense, we propose future research directions and emphasize the need for standardization of results to obtain time series and to allow comparison.

Also in this chapter, despite the low productivity of articles per year, we found that Brazil is the most productive country on this topic in relation to other tropical countries. The studies are distributed mainly in the Northern and Northeast regions, following the extension of mangroves along the Brazilian coast. While the estuarine systems of the Northern region are influenced by the annual freshwater discharge of the Amazon River, the Northeastern systems are influenced by periods of little or no freshwater discharge. In the latter, the Semiarid Coast of Brazil is characterized by the hypersaline condition in the mangroves, which negatively affects the ichthyoplankton community and the recruitment of species of economic interest. Therefore, chapters 02 and 03 fulfill this need, highlighting the distribution and structure of fish eggs and larvae in estuaries of the Semiarid Coast of Brazil in two extreme events: severe drought vs. heavy precipitation.

In Chapter 02, the hypersaline condition (sal. > 60) of the Piranji River estuary negatively affected the survival of fish eggs and larvae, and the use of this estuary as a nursery zone. The low density and diversity of the ichthyoplankton evidences a higher sensitivity of these organisms to extreme environmental changes, which leads to a lower survivability, difficulty for osmoregulation due to a high energy expenditure, and the use of the estuary only as a nursery area for few salinity-tolerant species. In fact, fish eggs and larvae were found in higher density in areas of lower salinity. Therefore, using the Piranji River estuary as a model, our conclusions show that possible extreme drought events can reduce the nursery function of mangroves for freshwater species and affect the fish stocks that are essential to the lifestyle of traditional communities.

Chapter 03 was conducted from seasonal sampling in the Pacoti River estuary, which is in a sustainable use conservation unit. Although the hypersaline condition is an existing factor in this system, the strong and sporadic rainfall throughout the year prevented salinity values from exceeding the adjacent sea salinity.

The distribution pattern of the ichthyoplankton found in Pacoti was of eggs occurring in the region closest to the mouth and larvae concentrated in the upstream region, greatly influenced by the river. Since the fish larvae are in more advanced stages of development and are already feeding, we can highlight that this region is used as a feeding area for the larvae, but trophic ecology studies of the environment need to be developed to test this hypothesis. Moreover, another interesting result is the absence of freshwater species in the system, to the detriment of estuarine and marine species. This may be explained by the reproductive strategies of freshwater species and ability to the larvae to avoid being flushed out to less productive or inhospitable areas that prevented sampling.

Also, at Pacoti, we observed the presence of Ctenophores during the dry season sampling, in the downstream zone. It is known that these organisms prey on fish eggs and larvae and compete with fish for food. In this area, the Ctenophore *M. leidyi* (A. agassiz 1865) was found in the study of Campelo *et al.* (2020). The interaction of this species with ichthyoplankton is unknown for the region and needs further studies to better understand the low densities of ichthyoplankton.

The results presented in the three chapters reinforce the importance of estuaries for ichthyoplankton but demonstrate the negative influence of extreme weather events on their survival. In addition, predation/competition with other organisms and the presence of pollutants in the estuarine environment are also factors that can influence the density and diversity of ichthyoplankton. In a context of the United Nations Decade of Ocean Science for Sustainable Development and the Goals of Sustainable Development, studies such as this demonstrates the importance of understanding and conserving coastal environments and collaborating with the management of marine resources.