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Cytogenetic profiles of two circumglobal snake mackerel species (Scombriformes: Gempylidae) from deep waters of the São Pedro and São Paulo Archipelago

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Chromosomal patterns are valuable tools in evolutionary approaches. Despite the remarkable expansion of fish cytogenetic data, they are still highly deficient concerning deep oceanic species, including the Gempylidae snake mackerels. The snake mackerels are important commercial species composed by meso- and bento-pelagic predators with very limited information available about their lifestyle and genetics patterns. This study presents the first chromosomal data of two circumglobal species of this family, *Ruvettus pretiosus* and *Promethichthys prometheus*, from the São Pedro and São Paulo Archipelago. Conventional analyses, chromosomal staining with base-specific fluorochromes, and fluorescence *in situ* hybridization (FISH) for mapping of repetitive DNA classes were used. Both species have 2n = 48 chromosomes, but they highly differ regarding the karyotype formula (FN = 50 and FN = 84). The 18S rDNA/Ag-NOR and the 5S rDNA sites have a syntenic bi-telomeric array in *R. pretiosus*, but an independent distribution in *P. prometheus*. The transposable elements are dispersed, while the microsatellites are also clustered in the centromeric and terminal regions of some chromosomes. It is noteworthy that despite the 2n conservation, a marked macro and microstructural diversifications, mainly mediated by pericentric inversions, differentiates the karyotypes of the species, pointing to a particular chromosomal trajectory of the gempylids among marine fish.

Keywords: Karyotype evolution, Pericentric inversion, Repetitive DNAs, Transposable elements, Microsatellites.

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Padrões cromossômicos são ferramentas valiosas em abordagens evolutivas. Apesar da notável expansão dos dados citogenéticos dos peixes, eles ainda são altamente deficientes para as espécies de águas oceânicas profundas, incluindo os membros da família Gempylidae. Espécies desta família são comercialmente importantes, compostas por predadores meso e bentopelágicos, cujas informações disponíveis sobre seu estilo de vida e padrões genéticos são muito limitadas. Este estudo apresenta os primeiros dados cromossômicos de duas espécies circumglobais desta família, *Ruvettus pretiosus* e *Promethichthys prometheus*, do Arquipélago de São Pedro e São Paulo. Foram utilizadas análises convencionais, coloração cromossômica com fluorocromos base-específicos e hibridização *in situ* por fluorescência (FISH) para o mapeamento de diferentes classes de DNA repetitivos. Ambas as espécies possuem 2n = 48 cromossomos, mas diferem significativamente quanto à fórmula cariotípica (FN = 50 e FN = 84). Os sítios 18S DNAr/Ag-RON e 5S DNAr têm um arranjo bi-telomérico sintênico em *R. pretiosus*, mas uma distribuição independente em *P. prometheus*. Os elementos transponíveis têm dispersão semelhante em ambas as espécies, enquanto os microssatélites estão agrupados nas regiões centroméricas e terminais de alguns cromossomos. Vale ressaltar que apesar da conservação do 2n basal dos Percomorpha, uma acentuada diversificação macro e microestrutural, mediada principalmente por inversões pericêntricas, diferencia os cariótipos das espécies, apontando para uma trajetória cromossômica particular dos gempilídeos entre os peixes marinhos.

Palavras-chave: Evolução cariotípica, Inversão pericêntrica, DNA repetitivo, Elementos transponíveis, Microssatélites.

INTRODUCTION

Deep-sea regions constitute the largest habitat of the planet (Haedrich, 1996; Hobday *et al*., 2011), and are the home of the most abundant vertebrates on the Earth, the mesopelagic fishes (Kaartvedt *et al*., 2012; Proud *et al*., 2018). Eco-evolutionary studies have shown that genomic signatures are associated with fish adaptation to depth environments (Gaither *et al*., 2018). However, although mesopelagic species may provide important models for differentiation and adaptation processes in deep waters, very little is still known about their life history, mainly due to the inaccessibility that such marine regions offer (Caiger *et al*., 2021).

Scombriformes are among the fish groups with remarkable diversification and specialization in mesopelagic or deep environments. They comprise the suborders Scombroidei and Stromateoidei, and three families for the former: Gempylidae, Trichiuridae, and Scombridae (Miya *et al*., 2013). Gempylidae, the snake mackerels, includes 16 genera and 26 species (Fricke *et al*., 2024). They are usually large and fast meso- and bento-pelagic predators (Nelson *et al*., 2016), and can be found in the tropical and subtropical zones of all oceans, at depths from 200 to 500 m (Nakamura, Parin, 1993).

Some Atlantic snake mackerels have a circumtropical occurrence, such as the oilfish *Ruvettus pretiosus* Cocco, 1833, a benthopelagic species that reach up to three meters in length and has a high commercial value (Viana *et al*., 2012). The species occurs in tropical, subtropical and temperate waters of all oceans, at depths of 100 to 1,500 m (Nakamura, Parin, 1993). Another species, the Roudi escolar *Promethichthys prometheus* (Cuvier, 1832), is distributed in tropical and warm temperate waters, at continental slopes around oceanic islands and submarine rises, at depths from 100 to 800 m (Schneider, 1990; Lorenzo, Pajuelo, 1999). Both species perform daily vertical migration, moving to shallower waters at night in search of food (Nakamura, Parin, 1993).

Phylogeographic and population aspects of Gempylidae are still largely unknown, but some studies indicate that they can achieve genetic homogenization even between distantly situated regions (Hüne *et al*., 2021). Despite increasing chromosomal data among marine fish, large gaps remain for pelagic (Soares *et al*., 2021) and mesopelagic species (Molina *et al*., 2024), among which the Atlantic Gempylidae species are included.

Besides to chromosomal diversification at the macrostructural level, the particular organization of repetitive sequences is decisory for understanding the evolutionary trends within a biological group. In eukaryotes, about 20 to 90% of the genome is composed of repetitive sequences (Mehrotra, Goyal, 2014), which include multigene families, mobile elements, and satellite DNAs (Biscotti *et al*., 2015). Their high dynamic nature (Mehrotra, Goyal, 2014; Garrido-Ramos, 2015) allows for useful biogeographic, phylogenetic, and populational analyses (Vicari *et al*., 2010; Cioffi *et al*., 2018; Amorim *et al*., 2018; Soares *et al*., 2021; Fernandes *et al*., 2021).

In this study we aimed to improve the knowledge of evolutionary processes within mesopelagic ecosystems, using Gempylidae fish as a model. Thus, it was performed the first cytogenetic-evolutionary investigation in two Atlantic species, *R. pretiosus* and *P. prometheus*, using conventional analyses, staining with base-specific fluorochromes, and fluorescence *in situ* hybridization (FISH) of six repetitive DNA sequences, including rDNAs, transposable elements, and microsatellites. These first results already allow us to infer about the chromosomal diversification in Gempylidae species and its correlation with other marine fish.

MATERIAL AND METHODS

Cytogenetic analyses were performed on 10 individuals (4 males and 6 females) of *Ruvettus pretiosus*, and 5 individuals (3 males and 2 females) of *Promethichthys prometheus* from deep waters of the Brazilian São Pedro and São Paulo archipelago (00°55'15"N 29°20'60"W), in the Mid-Atlantic region (Fig. 1). Mitotic chromosomes were obtained by short-term *in vitro* culture of kidney tissues (Gold *et al*., 1990) and by lymphocyte culture (Moorhead *et al*., 1960). Cell suspensions were dripped on slides covered with a hot water film (60°C), and stained with a 5% Giemsa solution diluted in phosphate buffer pH 6.8. Chromosomes were also analyzed after C-banding (Sumner, 1972), Silver nitrate impregnation (Howell, Black, 1980), and chromomycin (CMA_3) and 4'-6-diamino-2-phenylindole (DAPI) staining (Schweizer, 1980), to identify the heterochromatin distribution, the nucleolar organizer regions location and the chromosomal GC- and AT-rich regions, respectively.

Fluorescence *in situ* hybridization (FISH) was performed using 18S rDNA, 5S rDNA, the retroelement of *Xiphophorus* 3 (*Rex 3*) and transposable element (TE) of *Oryzias latipes*, number 2 (*Tol2*) as probes. The 5S rDNA (200 base pairs) and 18S rDNA (1400 bp) probes were obtained from the genomic DNA of *Rachycentron canadum* (Rachycentridae) via PCR, using the primers A 5'-TAC GCC CGA TCT CGT CCG ATC-3'/ B 5' GAG AGC GCT GGT ATG GCC AGC-3' (Pendás *et al*., 1994) and NS1 5'-GTA GTA ATA TGC TTG TCT C-3' / NS8 5'-TCC GCA GGT TCA CCT ACG GA-3' (White *et al*., 1990), respectively. *Rex3* and *Tol2* probes were obtained via PCR from the amplification of the *P. prometheus* DNA, using the primers *Rex* 3 F 5′ - CGG TGA TAA AGG GCA GCC GTC - 3′ and *Rex* 3 R 5′- TGG CAG ACN GTG GTG GTG - 3' (Volff *et al*., 1999, 2000) and 4F 5' - ATA GCT GAA GCT GCT CTG ATC - 3' and 4R 5' - CTC AAT ATG CTT CCT TAG G - 3' (Kawakami, Shima, 1999). The probes were labeled by nick translation (Roche®, Mannheim, Germany) with digoxigenin-11-dUTP, following the manufacturer's instructions (Roche®, Mannheim, Germany). The $d(CA)_{15}$ and $d(GA)_{15}$ oligonucleotides were directly labeled with Alexa-Fluor 555 (InvitrogenTM, Thermo Fisher Scientific, California, USA), at the 5' terminal position (Kubat *et al*., 2008). The FISH protocol was performed according to Pinkel *et al*. (1986).

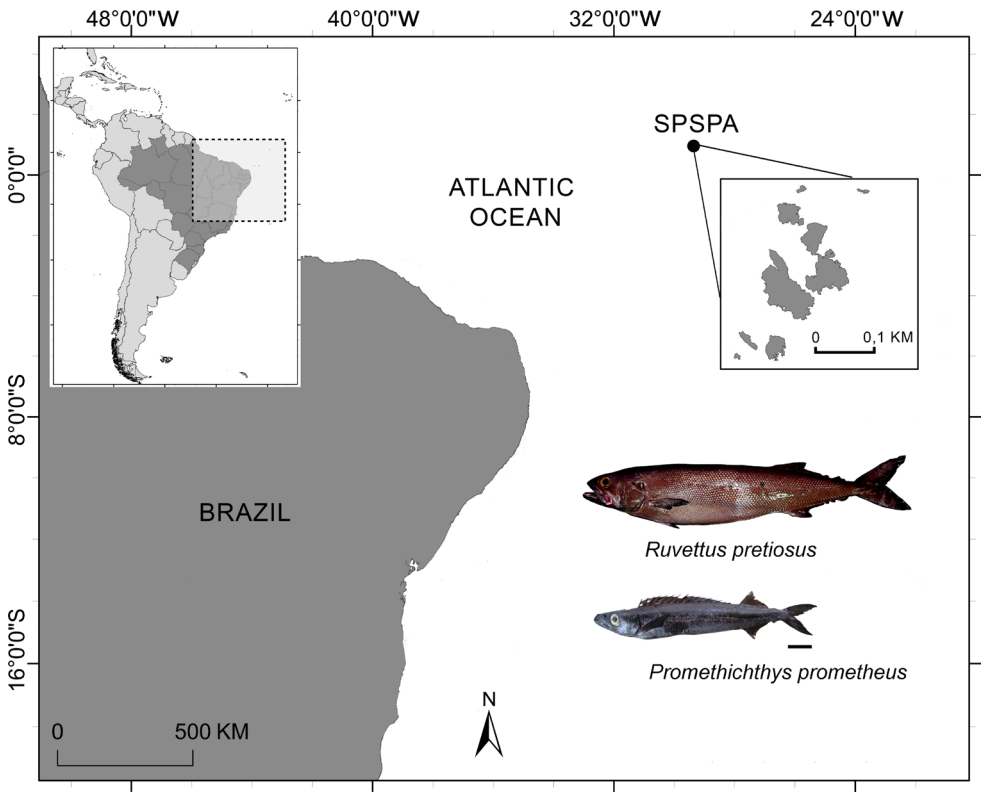

FIGURE 1 | South America map showing the geographic location of the São Pedro and São Paulo Archipelago, and the Gempylidae species analyzed in this study. Scale bar = 10 cm.

The best metaphases were photographed using an Olympus™BX51 epifluorescence microscope, coupled with an Olympus™DP73 digital image capture system (Olympus Corp., Tokyo, Japan). The images were compiled with CellSens v. 1.5 Imaging software (Olympus Corp.). Chromosomes were classified according to their arm ratios (AR) as metacentric (m: AR = 1.00–1.70), submetacentric (sm: AR = 1.71–3.00), subtelocentric (st: AR = 3.01–7.00), and acrocentric (a: AR > 7.01), according to Levan *et al*. (1964). The number of chromosome arms (Fundamental Number, FN) was obtained considering the m, sm, and st chromosomes with two arms, and the acrocentric ones with only one arm.

RESULTS

Promethichthys prometheus and *R. pretiosus* share 2n = 48 chromosomes but differ considerably in their karyotypic formulas. The karyotype of *R. pretiosus* is composed of 2 submetacentric and 46 acrocentric chromosomes (FN = 50), while *P. prometheus* has 34 submetacentric, 4 subtelocentric and 10 acrocentric chromosomes (FN = 86) (Fig. 2). The Ag-NOR site is located in the short arms of the only submetacentric pair (pair 1) in *R. pretiosus*, and the short arm of the sm pair 6 in *P. Prometheus*. In the two species, the Ag-NORs are associated with conspicuous heterochromatic blocks, which are the only chromosomal regions showing differential fluorescence patterns (CMA_3^+/$DAPI^-$). The heterochromatin is also preferentially located in the peri- and centromeric regions of the chromosomes in both species (Fig. 2).

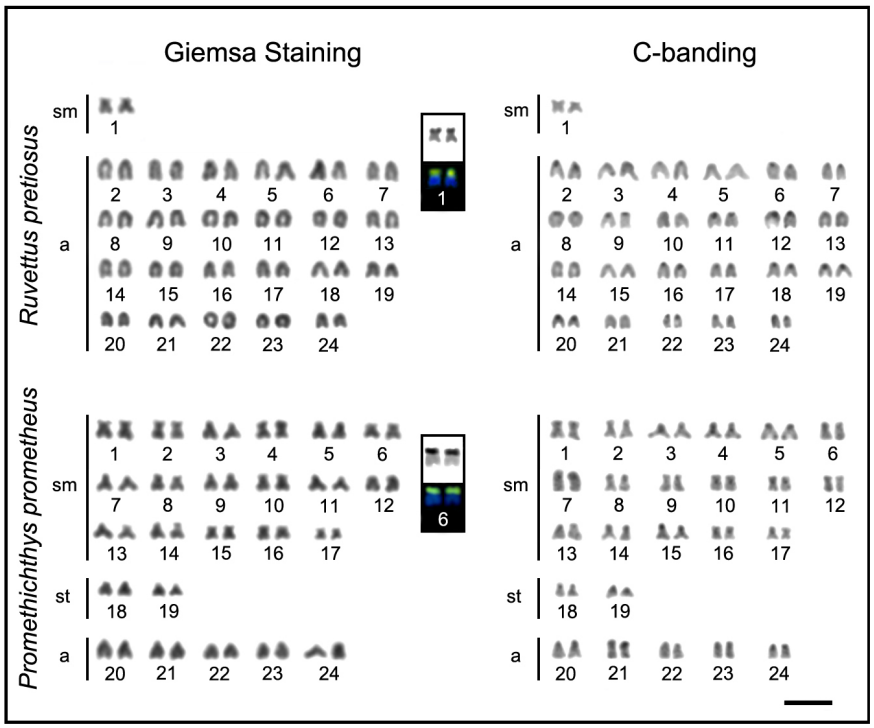

FIGURE 2 | Karyotypes of *Ruvettus pretiosus* and *Promethichthys prometheus*, under Giemsa staining and C-banding. The Ag-NORs sites and the correspondent CMA^+/$DAPI^-$ regions are highlighted in the boxes. Scale bar = 5µm.

The 18S rDNA hybridization signals are coincident with the Ag-NORs sites. However, while in *R. pretiosus* the 18S rDNA has a syntenic arrangement with the 5S in pair 1, in *P. prometheus* the 18S and 5S rDNAs have an independent location, the first in pair 6 and the second in pair 8 (Fig. 3). Furthermore, $(GA)_{15}$ and $(CA)_{15}$ microsatellites, and *Tol2* transposable element just have scattered signals on the chromosomes in *R. pretiosus* and *P. prometheus*. *Rex3* showed scattered signals on the chromosomes in *R. pretiosus* and *P. prometheus* besides accumulations in pericentromeric region of chromosome pairs 1, 2, 7, and 12 in *P. prometheus* (Figs. 3–4).

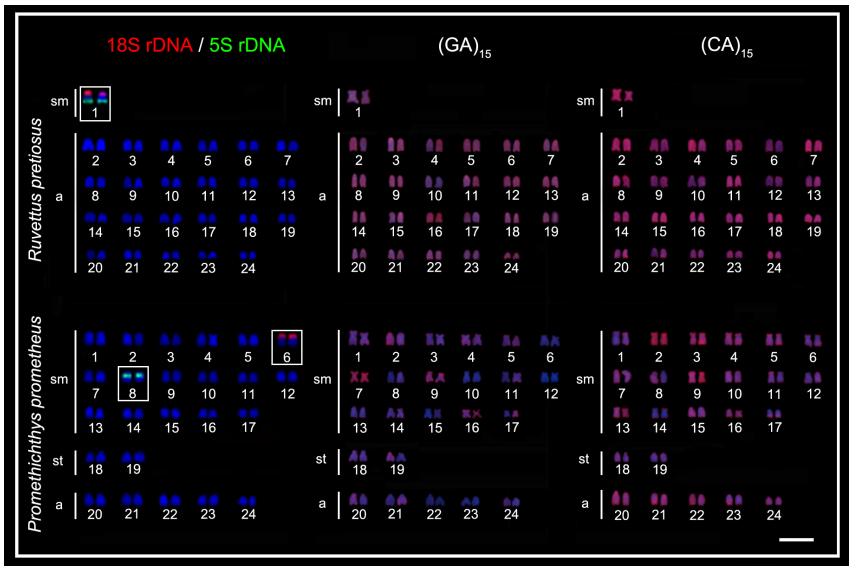

FIGURE 3 | Karyotypes of *Ruvettus pretiosus* and *Promethichthys prometheus* showing the distribution of the 18S rDNA (red), 5S rDNA (green) probes, and microsatellites $(GA)_{15}$ and $(CA)_{15}$ under fluorescence *in situ* hybridization. Scale bar = 5 µm.

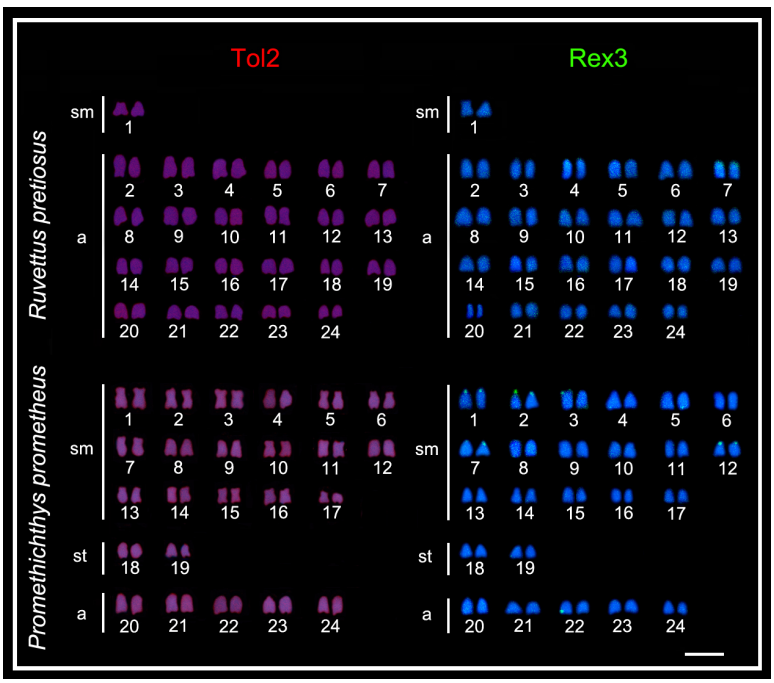

FIGURE 4 | Karyotypes of *Ruvettus pretiosus* and *Promethichthys prometheus* showing the distribution of the *Tol2* and *Rex3* elements in the chromosomes under fluorescence *in situ* hybridization. Scale bar = 5 µm.

DISCUSSION

Cytogenetic data have not been described for a significant number of marine fish yet, especially those whose access or management is difficult due to their large size, remote geographic distribution or very specific ecological habitats. All these attributes apply to Gempylidae species, usually living at great ocean depths. Therefore, this study provides the first information about this small and little-studied fish group.

Although sharing the same diploid number, 2n = 48, *R. pretiosus* and *P. prometheus* have diversified karyotypic structures. Their chromosomal number is considered a basal trait for Percomorpha fish (Galetti *et al*., 2000; Motta-Neto *et al*., 2019), and is also shared by 15 other Scombridae species, a sister family of Gempylidae also belonging to the Scombroidei clade (Arai, 2011; Soares *et al*., 2013). This symplesiomorphic condition is frequently found among Perciformes (Molina, 2007; Motta-Neto *et al*., 2019), indicating that other rearrangements, regardless of centric fissions, have played an important role in the karyotypic evolution of this fish group. However, in contrast to other Scombroidei fish, the two analyzed Gempylidae species have a very distinctive FN, because of their divergent karyotypic diversification. Thus, while *R. pretiosus* has all acrocentric chromosomes, except for a single sm pair (FN = 50), *P. prometheus*, has almost two-armed chromosomes (FN = 86). Such expressive differentiation is likely due to pericentric inversions, the most frequent chromosomal rearrangements in Percomorpha (Galetti *et al*., 2006), but not excluding *a priori* other rearrangements that may have acted in the shuffling of syntenic regions of the chromosomes. Pericentric rearrangements are seen as important tools for local adaptations (Wellenreuther, Bernatchez, 2018), and have been correlated with such processes in several fish groups (Matschiner *et al*., 2022). Therefore, it is also likely that they are acting in the adaptation of some species of Gempylidae to deep marine environments.

Biological factors, such as their dispersive potential, can influence the karyotypic evolution in marine fishes (Molina, Galetti, 2004; Sena, Molina, 2007; Soares *et al*., 2021). In fact, the dispersive capacity, including the transposition of geographic barriers (Fernandes *et al*., 2021), and variable ecological abilities, may minimize the genetic structuring and reduce the fixation of chromosomal rearrangements, while opposite characteristics may facilitate them (Molina, 2007; Motta-Neto *et al*., 2019). Phylogeographic data are still unavailable to *R. pretiosus* and *P. prometheus*, but their diversified karyotypic patterns are apparently in concordance with their dispersive potentials. Phylogenetically, *R. pretiosus* is a more basal species (Miya *et al*., 2013), with extensive distribution in tropical, subtropical, and temperate deep waters of all oceans (Nakamura, Parin, 1993), indicating a bigger dispersive potential. Accordingly, it presents a more conserved karyotypic structure. On the other hand, *P. prometheus*, which is included in a more recent divergent group among the snake mackerels (Miya *et al*., 2013), and with a circumglobal distribution, but not in the eastern Pacific (Nakamura, Parin, 1993), presents a significantly differentiated karyotype.

The distribution of the rDNA sequences shows that microstructural divergences also occur between the two species. Outstandingly, the 18S and 5S rDNA sites are localized in two different chromosome pairs in *P. prometheus*, while in *R. pretiosus* these sequences are localized in the telomeric regions of the same chromosome pair. Although contiguous syntenic arrays have been sporadically reported for some Percomorpha species (Nirchio

et al., 2009; Amorim *et al*., 2016; Motta-Neto *et al*., 2019), the bi-telomeric rDNA organization is a rare array in marine fish.

In fish, microsatellites are frequently associated with TEs (Costa *et al*., 2015; Gouveia *et al*., 2017), and can show highly variable accumulation patterns (Cioffi *et al*., 2012; Lima-Filho *et al*., 2014). However, in *P. prometheus* and *R. pretiosus*, the $(CA)_{15}$ and $(GA)_{15}$ repeats are homogeneously dispersed in chromosomes, with some centromeric and terminal clusters in a few chromosomes. The distributions of the microsatellites repeats and *Tol2* elements show no significant differences between both species. Except by the accumulation in few chromosomes of *P. prometheus*, the *Rex3* sequences are disperse on the chromosomes of both species, in contrast with other marine species, in which they are visibly clustered (Ferreira *et al*., 2011; Costa *et al*., 2013, 2014, 2015). Despite TEs are recognized as sources of chromosomal instability, favoring karyotypic differentiation (Lonnig, Saedler, 2002; Shao *et al*., 2019), were not evidenced complex arrays involving the analyzed TEs with repetitive DNA sequences, such rDNA regions, or microsatellites, suggesting their less direct participation on karyotype divergence of these snake mackerel species.

These chromosomal data, now recorded for the first time for Gempylidae, indicate clear macrostructural differences between the two investigated species, in contrast to the conservative trend that occurs in its phylogenetically close and cytogenetically more studied Scombridae family (Soares *et al*., 2013). It is known that high intrafamilial diversification is a common scenario in some reef fish groups (Molina *et al*., 2014; Getlekha *et al*., 2016), but hitherto unknown in deep-sea fish such as Gempylidae.

The vast areas of the Mid-Atlantic Ridge and other global Mid-Oceanic Ridges systems, used as spawning grounds for deep-sea fish, may have a strong influence on their genetic structure (Sutton *et al*., 2007). In fact, the common strategy of the vertically migrating mesopelagic species in releasing eggs near the surface (Gjøsæter, Tilseth, 1988; Flynn, Paxton, 2012), amplifies their dispersive potential, making them good models for investigating chromosomal evolution in marine environments.

Regions with little to no coverage of mesopelagic fish research include the South Atlantic, large parts of Indo-Pacific region and some polar environments (Caiger *et al*., 2021). The lack of knowledge of the characteristics of deep pelagic species constitutes a challenge for the conservation of global oceanic biodiversity (Sutton *et al*., 2017). The cytogenetic patterns and life history of pelagic fish are beginning to be better analyzed. Undoubtedly, this will be an important step towards better understanding our rich biodiversity and its correlation with the environment where it lives.

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REFERENCES

- **Arai K.** Fish karyotypes: A check list. New York: Springer; 2011.
- **Biscotti MA, Olmo E, Heslop-Harrison JS.** Repetitive DNA in eukaryotic genomes. Chromosome Res. 2015; 23:415–20. https://doi.org/10.1007/s10577-015-9499-z_
- **Caiger PE, Lefebve LS, Llopiz JK.** Growth and reproduction in mesopelagic fishes: a literature synthesis. ICES J Mar Sci. 2021; 78(3):765–81. https://doi.org/10.1093/icesjms/fsaa247
- **Cioffi MB, Kejnovsky E, Marquioni V, Poltronieri J, Molina WF, Diniz D *et al*.** The key role of repeated DNAs in sex chromosome evolution in two fish species with ZW sex chromosome system. Mol Cytogenet. 2012; 5:28. https://doi.org/10.1186/1755-8166-5-28
- **Cioffi MB, Moreira-Filho O, Ráb P, Sember A, Molina WF, Bertollo LAC.** Conventional cytogenetic approaches - Useful and indispensable tools in discovering fish biodiversity. Curr Genet Med Rep. 2018; 6:176–86. https://doi.org/10.1007/s40142-018-0148-7
- **Costa GWWF, Cioffi MB, Bertollo LAC, Molina WF.** Transposable elements in fish chromosomes: A study in the marine Cobia species. Cytogenet Genome Res. 2013; 141(2–3):126–32. https://doi.org/10.1159/000354309
- **Costa GWWF, Cioffi MB, Bertollo LAC, Molina WF.** Unusual dispersion of histone repeats on the whole chromosomal complement and their colocalization with ribosomal genes in *Rachycentron canadum* (Rachycentridae, Perciformes). Cytogenet Genome Res. 2014; 144(1):62–67. https://doi.org/10.1159/000366301
- **Costa GWWF, Cioffi MB, Bertollo LAC, Molina WF.** Structurally complex organization of repetitive DNAs in the genome of Cobia (*Rachycentron canadum*). Zebrafish. 2015; 12(3):215–20. https://doi.org/10.1089/zeb.2014.1077
- **Fernandes MA, Cioffi MB, Bertollo LAC, Costa GWWF, Motta-Neto CC, Borges AT *et al*.** Evolutionary tracks of chromosomal diversification in surgeonfishes (Acanthuridae: *Acanthurus*) along the world's biogeographic domains. Front Genet. 2021; 12:760244. https://doi.org/10.3389/fgene.2021.760244
- **Ferreira DC, Oliveira C, Foresti F.** Chromosome mapping of retrotransposable elements *Rex1* and *Rex3* in three fish species in the subfamily Hypoptopomatinae (Teleostei, Siluriformes, Loricariidae). Cytogenet Genome Res. 2011; 132(1–2):64–70. https://doi.org/10.1159/000319620
- **Flynn AJ, Paxton JR.** Spawning aggregation of the lanternfish *Diaphus danae* (family Myctophidae) in the north-western Coral Sea and associations with tuna aggregations. Mar Freshw Res. 2012; 63(12):1255–71. https://doi.org/10.1071/MF12185
- **Fricke R, Eschmeyer WN, Fong JD.** Eschmeyer's catalog of fishes: Genera/species by family/subfamily [Internet]. San Francisco: California Academy of Science; 2024. Available from: http://researcharchive.calacademy.org/research/ichthyology/catalog/SpeciesByFamily.asp
- **Gaither MR, Gkafas GA, Jong M, Sarigol F, Neat F, Regnier T *et al*.** Genomics of habitat choice and adaptive evolution in a deep-sea fish. Nat Ecol Evol. 2018; 2:680–87. https://doi.org/10.1038/s41559-018-0482-x
- **Galetti PM Jr., Aguilar CT, Molina WF.** An overview on marine fish cytogenetics. Hydrobiologia. 2000; 420:55–62. https://doi.org/10.1023/A:1003977418900
- **Galetti PM Jr., Molina WF, Affonso PRAM, Aguilar CT.** Assessing genetic diversity of Brazilian reef fishes by chromosomal and DNA markers. Genetica. 2006; 126:161–77. https://doi.org/10.1007/s10709-005-1446-z
- **Garrido-Ramos MA.** Satellite DNA in plants: more than just rubbish. Cytogenet Genome Res. 2015; 146(2):153–70. https://doi.org/10.1159/000437008
- **Gjøsæter J, Tilseth S.** Spawning behavior, egg and larval development of the myctophid fish *Benthosema pterotum*. Mar Biol. 1988; 98:1–06. https://doi.org/10.1007/BF00392652
- **Gold JR, Li YC, Shipley NS, Powers PK.** Improved methods for working with fish chromosomes with a review of metaphase chromosome banding. J Fish Biol. 1990; 37(4):563–75. https://doi.org/10.1111/j.1095-8649.1990.tb05889.x

- **Gouveia JG, Wolf IR, Vilas-Boas LA, Heslop-Harrison JS, Schwarzacher T, Dias AL.** Repetitive DNA in the catfish genome: rDNA, microsatellites, and Tc1-Mariner transposon sequences in *Imparfinis* species (Siluriformes, Heptapteridae). J Heredity. 2017; 108(6):650–57. https://doi.org/10.1093/jhered/esx065
- **Haedrich RL.** Deep-water fishes: evolution and adaptation in the earth's largest living spaces. J Fish Biol. 1996; 49:40–53. https://doi.org/10.1111/j.1095-8649.1996.tb06066.x
- **Hobday AJ, Game ET, Grantham HS, Richardson AJ.** Missing dimension: Conserving the largest habitat on Earth: protected areas in the pelagic ocean. Marine Protected Areas: A Multidisciplinary Approach. Cambridge, UK: Cambridge University Press, 2011. p.347–72. https://doi.org/10.1017/CBO9781139049382.019
- **Howell WM, Black DA.** Controlled Silver-staining of nucleolus organizer regions with a protective colloidal a 1-step method. Experientia. 1980; 36:1014–15. https://doi.org/10.1007/BF01953855
- **Hüne M, Oyarzún PA, Reys P, Gladys R, Montecinos M.** Phylogeographic analysis of *Thyrsites atun* (Perciformes: Gempylidae) reveals connectivity between fish from South Africa and Chile. Mar Biol Res. 2021; 17(4):401–13. https://doi.org/10.1080/17451000.2021.1967994
- **Kaartvedt S, Staby A, Aksnes DL.** Efficient trawl avoidance by mesopelagic fishes causes large underestimation of their biomass. Mar Ecol Prog Ser. 2012; 456:1–06.
- **Kawakami K, Shima A.** Identification of the *Tol2* transposase of the medaka fish *Oryzias latipes* that catalyzes excision of a non-autonomous *Tol2* element in zebrafish *Danio rerio*. Gene 1999; 240(1):239–44. https://doi.org/10.1016/s0378-1119(99)00444-8
- **Kubat Z, Hobza R, Vyskot B, Kejnovsky E.** Microsatellite accumulation on the Y chromosome in *Silene latifolia*. Genome. 2008; 51(5):350–56. https://doi.org/10.1139/G08-024
- **Levan A, Fredga K, Sandberg AA.** Nomenclature for centromeric position on chromosomes. Hereditas. 1964; 52(2):201–20. https://doi.org/10.1111/j.1601-5223.1964.tb01953.x
- **Lima-Filho PA, Bertollo LAC, Cioffi MB, Costa GWWF, Molina WF.** Karyotype divergence and spreading of 5S rDNA sequences between genomes of two species: Darter and emerald gobies (*Ctenogobius*, Gobiidae). Cytogenet Genet Res. 2014; 142(3):197–203. https://doi.org/10.1159/000360492
- **Lonnig W-E, Saedler H.** Chromosome rearrangements and transposable elements. Annu Rev Genet. 2002; 36:389–410. https://doi.org/10.1146/annurev.genet.36.040202.092802
- **Lorenzo JM, Pajuelo JG.** Biology of a deep benthopelagic fish, roudi escolar *Promethichthys prometheus* (Gempylidae) off the Canary Islands. Fish Bull. 1999; 97:92–99.
- **Matschiner M, Barth JMI, Tørresen OK, Star B, Baalsrud HT, Brieuc MSO *et al*.** Supergene origin and maintenance in Atlantic cod. Nat Ecol Evol. 2022; 6:469–81. https://doi.org/10.1038/s41559-022-01661-x
- **Mehrotra S, Goyal V.** Repetitive sequences in plant nuclear DNA: types, distribution, evolution and function. Genom Proteom Bioinform. 2014; 12:164–71. https://doi.org/10.1016/j.gpb.2014.07.003
- **Miya M, Friedman M, Satoh TP, Takeshima H, Sado T, Iwasaki W *et al*.** Evolutionary origin of the Scombridae (Tunas and Mackerels): Members of a Paleogene adaptive radiation with 14 other pelagic fish families. PLoS ONE. 2013; 8(9):e73535. https://doi.org/10.1371/journal.pone.0073535
- **Molina WF.** Chromosome changes and stasis in marine fish groups. In: Pisano E, Ozouf-Costaz C, Foresti F, Kapoor BG, editors. Fish cytogenetic. Boca Raton, USA: CRC Press, 2007. p.69–110.
- **Molina WF, Amorim KDJ, Silva SAS, Cioffi MB, Bertollo LAC, Soares RX *et al*.** Karyotype evolutionary diversification in marine fishes. First classical and molecular cytogenetic data on four Atlantic species. N Z J Mar Freshw Res. 2024; 1–13. https://doi.org/10.1080/00288330.2024.2328138
- **Molina WF, Galetti Jr. PM.** Karyotypic changes associated to the dispersive potential on Pomacentridae (Pisces, Perciformes). J Exp Mar Bio Ecol. 2004; 309(1):109–19. https://doi.org/10.1016/j.jembe.2004.03.011

- **Molina WF, Martinez PA, Bertollo LAC, Bidau CJ.** Evidence for meiotic drive as an explanation for karyotype changes in fishes. Mar Genomics. 2014; 15:29–34. https://doi.org/10.1016/j.margen.2014.05.001
- **Moorhead PS, Nowell PC, Mellmam WJ, Battips DM, Hungerford DA.** Chromosome preparations of leukocytes cultured from human peripheral blood. Exp Cell Res. 1960; 20:613–16.
- **Nakamura I, Parin NV.** FAO species catalogue. Snake mackerels and cutlassfishes of the world (families Gempylidae and Trichiuridae). FAO Fisheries Synopsis, 1993; 15:1–136.
- **Nelson JS, Grande TC, Wilson MVH.** Fishes of the World. 5th Edition, John Wiley and Sons: Hoboken; 2016.
- **Pendás AM, Morán P, García-Vázquez E.** Organization and chromosomal location of the major histone cluster in brown trout, Atlantic salmon and rainbow trout. Chromosoma. 1994; 103:147–52. https://doi.org/10.1007/BF00352324
- **Pinkel D, Straume T, Gray JW.** Cytogenetic analysis using quantitative, high sensitivity, fluorescence hybridization. Proc Natl Acad Sci. 1986; 83(9):2934–38. https://doi.org/10.1073/pnas.83.9.2934
- **Proud R, Handegard NO, Kloser RJ, Cox MJ, Brierley AS.** From siphonophores to deep scattering layers: uncertainty ranges for the estimation of global mesopelagic fish biomass. ICES J Mar Sci. 2018; 76(3):718–33. https://doi.org/10.1093/icesjms/fsy037
- **Schweizer D.** Simultaneous fluorescent staining of R bands and specific heterochromatic regions (DA-DAPI bands) in human chromosomes. Cytogenet Cell Genet. 1980; 27(2–3):190–93. https://doi.org/10.1159/000131482
- **Shao F, Han M, Peng Z.** Evolution and diversity of transposable elements in fish genomes. Sci Rep. 2019; 9:15399. https://doi.org/10.1038/s41598-019-51888-1
- **Soares RX, Bertollo LAC, Costa GWWF, Molina WF.** Karyotype stasis in four Atlantic Scombridae fishes: Mapping of classic and dual-color FISH markers on chromosomes. Fish Sci. 2013; 79:177–83. https://doi.org/10.1007/s12562-013-0602-0
- **Soares RX, Costa GWWF, Cioffi MB, Bertollo LAC, Motta-Neto CC, Molina WF.** Molecular cytogenetics insights in two pelagic big-game fishes in the Atlantic, the tarpon, *Megalops atlanticus* (Elopiformes: Megalopidae), and the sailfish, *Istiophorus platypterus* (Istiophoriformes: Istiophoridae). Neotrop Ichthyol. 2021; 19(2):e210007. https://doi.org/10.1590/1982-0224-2021-0007
- **Sumner AT.** A simple technique for demonstrating centromeric heterochromatin. Exp Cell Res. 1972; 75(1):304–06. https://doi.org/10.1016/0014-4827(72)90558-7
- **Sutton TT, Clark MR, Dunn DC, Halpin PN, Rogers AD, Guinotte J *et al.*** A global biogeographic classification of the mesopelagic zone. Deep Sea Res. 2017; 126:85–102. https://doi.org/10.1016/j.dsr.2017.05.006
- **Sutton TT, Porteiro FM, Horne J, Anderson CIH.** Meso- and bathypelagic fish interactions with seamounts and mid-ocean ridges. Marine & Environmental Sciences Faculty Proceedings, Presentations, Speeches, Lectures; 2007.
- **Vicari MR, Nogaroto V, Noleto RB, Cestari MM, Cioffi MB, Almeida MC *et al.*** Satellite DNA and chromosomes in Neotropical fishes: applications and perspectives. J Fish Biol. 2010; 76(5):1094–116. https://doi.org/10.1111/j.1095-8649.2010.02564.x
- **Volff J-N, Körting C, Schartl M.** Multiple lineages of the non-LTR retrotransposon *Rex1* with varying success in invading fish genomes. Mol Biol Evol. 2000; 17(11):1673–84. https://doi.org/10.1093/oxfordjournals.molbev.a026266
- **Volff J-N, Körting C, Sweeney K, Schartl M.** The non-LTR retrotransposon *Rex3* from the fish *Xiphophorus* is widespread among teleosts. Mol Biol Evol. 1999; 16(11):1427–38. https://doi.org/10.1093/oxfordjournals.molbev.a026055
- **Wellenreuther M, Bernatchez L.** Eco-evolutionary genomics of chromosomal inversions. Trends Ecol Evol. 2018; 33(6):427–40. https://doi.org/10.1016/j.tree.2018.04.002

- **White TJ, Bruns T, Lee S, Taylor J.** Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In: PCR Protocols: a guide to methods and applications. Academic Press Inc., 1990. p.315–22.

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ETHICAL STATEMENT

Collections had the authorization of the Instituto Chico Mendes de Conservação da Biodiversidade (ICMBio), Sistema de Autorização e Informação em Biodiversidade (SISBIO Licenses Nº 19135–1, 131360–1 and 27027–2), and Sistema Nacional de Gestão do Patrimônio Genético e do Conhecimento Tradicional Associado (SISGEN). All experiments followed ethical protocols approved by the Animal Ethics Committee of the Universidade Federal do Rio Grande do Norte (Protocol 44/2015).

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The authors no declare competing interests.

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