

Article

# Geographic Range Expansion and Taxonomic Notes of the Shortfin Neoscopelid *Neoscopelus* cf. *microchir* (Myctophiformes: Neoscopelidae) in the North-Eastern Atlantic

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**Abstract:** One specimen of the shortfin neoscopelid *Neoscopelus microchir* Matsubara, 1943, has been recorded for the first time on the Porcupine Bank, southwestern Ireland, providing a new northern limit of distribution for the eastern Atlantic. Morphometric and meristic parameters confirm the taxonomic identification. However, DNA barcoding shows deficiencies in current taxonomy and the potential occurrence of cryptic species. On this basis, the specimen is cautiously reported as *Neoscopelus* cf. *microchir* pending a taxonomic revision of the genus.

**Keywords:** NE Atlantic; deep-water fishes; seamount; cryptic species; distribution; blackchins



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## 1. Introduction

The order Myctophiformes includes the family Neoscopelidae Jordan, 1901 and Myctophidae Gill, 1893, both composed mainly of luminous, pelagic or benthopelagic fishes that occupy deep-sea habitats in all oceans [1]. The family Neoscopelidae is represented by six species in three genera: *Neoscopelus* Johnson, 1863, *Scopelengys* Alcock, 1890 and *Solvomere* Miller, 1947. The species of the genus *Neoscopelus* are characterized by being the only components of this family that possess photophores. The genus is composed of three species, two of them of circumglobal distribution, the large-scaled lantern fish *Neoscopelus macrolepidotus* Johnson, 1863 and the shortfin neoscopelid *Neoscopelus microchir* Matsubara, 1943, and the other the spangleside neoscopelid *Neoscopelus porosus* Arai, 1969, restricted to the Indo-West Pacific.

*Neoscopelus microchir* is a medium-sized benthopelagic marine species, up to 37 cm in total length (TL), inhabiting continental and island slopes and seamounts at depths from 250 to 896 m [2,3]. It is widespread in tropical and warm temperate areas of the Atlantic-Indo-West Pacific, but not in the eastern Pacific [2]. In the western Atlantic it is found in the Straits of Florida and northern Gulf of Mexico and off Jamaica and Honduras [4], whereas in the eastern Atlantic it is reported from the Galicia Bank (NW Spain) to Morocco, including the Macaronesian islands, and South Africa [3].

Although the fish fauna of the Bank is relatively well studied [5,6], the annual bottom trawl survey carried out in this area and a greater effort towards the study of marine biodiversity have led to a better understanding the Bank's ichthyofauna. It is in this context that several fish species have been recorded on the Bank for the first time, as is the case with *Gadella maraldi* (Risso, 1810) [7]; *Bellotia apoda* Giglioli, 1883 [8]; *N. macrolepidotus* [9];

*Gaidropsarus granti* (Regan, 1903) and *Poecilopsetta beanii* (Goode, 1881) [10], most of them constituting new northern limits of distribution in the east Atlantic. The aim of this study is to provide a new northernmost limit of the distribution of *N. microchir* from the northeast Atlantic based on morphological and molecular characteristics and, in view of the results obtained, to discuss its current taxonomic status.

## 2. Materials and Methods

The Porcupine Bank (ICES Divisions 7c and 7k) is located in the northeast Atlantic, 200 km off the west coast of Ireland. It extends from a depth of 150 m to 4000 m of the abyssal plain, forming a seamount-like structure, which is connected by a narrow strip to the continental shelf. A Spanish bottom trawl research survey has been carried out annually since 2001 in the Bank, on board the R/V Vizconde de Eza, to monitor changes in the distribution and relative abundance of fish and fish assemblages; and the biological parameters of commercial fish species, for stock assessment purposes. The survey covers an area that extends from longitude 12° W to 15° W and from latitude 51° N to 54° N, following the standard methodology for the ICES IBTS North Eastern Atlantic Surveys.

In September–October 2021, during the 2021 Spanish Bottom Trawl Survey on the Porcupine Bank (SP-PORC-Q3), a total of 94 bottom trawls of 20 min duration were made between 196 and 1484 m depth using a Baca-GAV 39/52 with a cod-end mesh size of 20 mm.

In one of the trawls, a specimen that was preliminarily identified on board as *N. microchir* was caught and preserved frozen. Subsequent to the scientific survey, identification to species level was confirmed following Hulley [11]. After removing tissue samples for molecular analysis, which were stored at  $-28^{\circ}\text{C}$ , the specimen was preserved in 70% ethanol and deposited in the fish collection of the Museo Luis Iglesias de Ciencias Naturais in Santiago de Compostela (MHNUSC) under reference number MHN USC 25200.

A dot distribution map of the species was created based on georeferenced data contained in the GBIF and OBIS online marine biogeographic databases [12,13].

DNA was obtained from a sample of muscle tissue to sequence the standard 5' barcoding region of the mitochondrial *COI* gene, using the primer cocktail C\_FishF1t1-C\_FishR1t1 [14], following procedures described elsewhere [15]. A 652-nucleotides-long sequence was deposited in the GenBank and BOLD repositories under accession numbers ON774728 and PORCU026-22, respectively. The taxonomic status of the specimen was explored conducting a Neighbor-Joining analysis using MEGA version 11 [16]. All information regarding this specimen as well as its DNA barcode, image, place of capture and other complementary data are available from the "Fishes of the Porcupine Bank" project (code PORCU) in the Barcoding of Life Database (BOLD, <http://www.boldsystems.org>, accessed on 15 June 2022).

A further 24 DNA barcodes with the same species name were retrieved from the BOLD and GenBank databases, with which an alignment including the sequence of the specimen captured in Porcupine was performed.

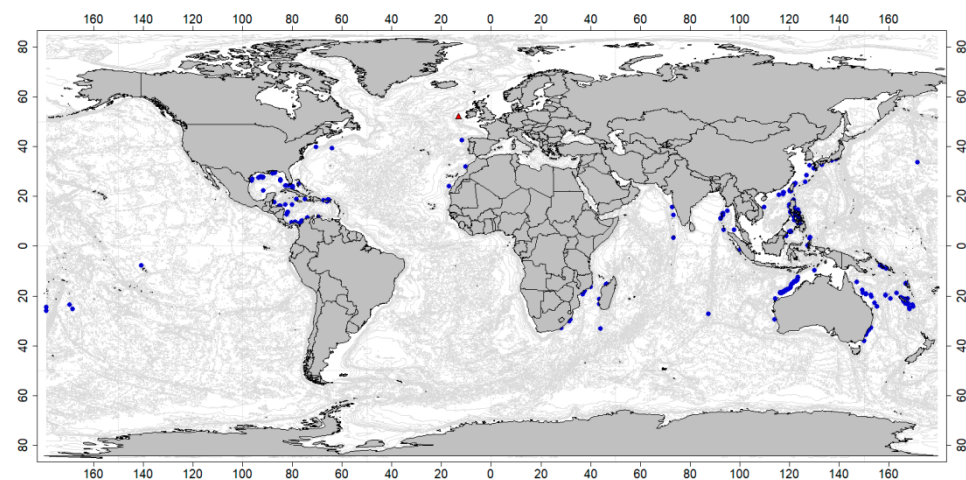
## 3. Results

On 20 September 2021, a specimen of *N. microchir* of 223.2 mm TL and 106 g of weight (Figure 1) was caught with bottom trawl gear in the Porcupine Bank.

A map showing the locations of the species worldwide shows that the present record at Porcupine Bank, 51.9796° N,  $-13.2322^{\circ}$  W, at 705 m depth (Figure 2) is the northernmost for the species in the eastern Atlantic and, as far as we know, worldwide.



**Figure 1.** *Neoscopelus microchir* MHN USC 25200, 223.2 mm TL, caught in the Porcupine Bank, southwestern Ireland (Photo: Francisco Baldó).



**Figure 2.** Distribution map of *Neoscopelus microchir* showing georeferenced location points of this species reported by GBIF and OBIS (blue dots). The red triangle mark represents the new location on the Porcupine Bank.

The main morphometric and meristic characteristics are presented in Table 1.

Regarding molecular taxonomy, the analysis of the distribution of paired differences among the 25 DNA barcodes using the Automatic Barcode Gap Discovery (ABGD) algorithm [17] revealed a barcode gap at a  $p$ -distance of 1.8% between intraspecific and interspecific diversities and a partition of the sequences into four groups or operational taxonomic units (OTU, i.e., species).

The taxonomic cladogram obtained by a Neighbor-Joining analysis (Figure 3) displayed a complete correspondence between the OTUs defined by the ABGD clustering method and the BINs present in the BOLD database, which designate OTUs through the RESL (Refined Single Linkage) algorithm, which applies a consensus threshold value of 2.2% of nucleotide differences in uncorrected pairwise distance ( $p$ -distance) as sequence divergence for OTU designation [18]. With this arrangement, the mean distances within each OTU varied in the percentage range 0.24–0.46, and those between OTUs ranged from 2.65% to 8.96%. The cladogram also allows for some consistency in relation to the geographical distribution of putative species.



**Figure 3.** Neighbor-Joining cladogram of the COI marker of *Neoscopelus microchir* specimens. There were 652 positions in the final sequence alignment. The numbers at the nodes indicate the percentage of tree resampling per bootstrap. The scale indicates the number of substitutions per nucleotide. The sequence of the specimen captured in Porcupine Bank is marked in bold. The correspondence between OTUs and BOLD BINs is also shown.

**Table 1.** Measurements and counts of specimen of *Neoscopelus microchir* MHN USC 25200 from Porcupine Bank and compared with the previous records from the Galicia Bank [19]. % LS: percentage of each measurement with respect to the Standard length (LS).

	Length (mm)	% LS	% LS [19]
Total length (TL)	223.2		-
Standard length (LS)	178.3		150–259 mm
Head length (LH)	53.8		29.7–34.5 mm
Preorbital length	12.4	30.2	6–9.4
Eye diameter	12.9	7	6.7–8.8
Postorbital length	28.5	7.2	14.3–18.1
Interorbital distance	18.3	16	6.6–8.6
Maxillar length	26.9	10.3	14–19.6
Predorsal length	73.4	15.1	41.4–48
Dorsal fin base length	28.7	41.2	13.5–18.3
Preadipose length	145.2	16.1	78.3–84.3
Preal length	133	81.4	73.1–79
Anal fin base length	18.2	74.6	8–11
Prepectoral length	53.4	10.2	-
Pectoral length	55.7	29.9	26.7–34.9
Prepelvic length	74.6	31.2	41.8–47.5
Pelvic length	22.3	41.8	12.4–18.3
Caudal depth	18.1	12.5	-
Body depth	42.1	10.2	18.9–28.1
Body width	25.9	23.6	10.6–14.9
No. of dorsal fin rays	14		13–14
No. of anal fin rays	12		10–11
No. of pectoral fin rays	17		16–18
No. of ventral fin rays	9		9
Branchiostegal rays	9		9
Gill rakers	3 + 1 + 10		3 + 1 + 10–11
LO photophores	23		20–24
Is photophores	9		8–9

#### 4. Discussion

Two *Neoscopelus* species, *N. macrolepidotus* and *N. microchir*, have been reported in the Atlantic Ocean [4,11]. The number of photophores in the LO series and the number of gill rakers are diagnostic features that distinguish one species from the other [20]. The

photophores in the LO series are 20–22, reaching to about the end of the anal fin base in *N. microchir* for 12–14 photophores that do not reach the anal fin origin in *N. macrolepidotus*, whereas *N. microchir* has 12–14 gill rakers in the first arch for only 10–12 in *N. macrolepidotus* [11,21]. The specimen examined, with 23 photophores in the LO series reaching the end of the anal fin base and 14 gill rakers, would confirm identification as *N. microchir*.

However, in this case, molecular taxonomy showed discrepancies with the morphological identification. The results indicate the existence of four *Neoscopelus* lineages. Although DNA barcoding is predominantly a tool for species identification, in this case we found profound genetic divergences that could simply be due to misidentification of specimens or involve cryptic or unrecognized speciation events. Indicators of critical speciation have been proposed before, such as the 10× rule, according to which barcoded individuals are flagged as possible critical species if they diverge 10 or more times from their group in intraspecific distances [22], which our data seem to reflect. The molecular results confirm the great complexity of the systematics of the genus *Neoscopelus* and show that DNA barcoding can be a useful tool to address these complex questions.

This taxonomic inconsistency has already been observed previously. Gaither et al. [23] noted that sequences of *N. microchir* in BOLD fall into three BINs, indicating divergent and overlapping lineages. Teramura et al. [24] found that *N. microchir* was divided into four lineages, and two lineages are sympatrically distributed in the northwestern Pacific Ocean and South Africa.

The possible presence of cryptic species for Atlantic specimens is also suggested in other taxonomic works. In a recent key to the identification of eastern central Atlantic fishes, Hulley and Paxton [2] consider the family to contain seven species rather than the generally recognized six, and assign the distinctive characters of *N. microchir* to a new species, reported as *Neoscopelus* n. sp., although not yet published. *Neoscopelus microchir* was originally described from the Pacific Ocean by Matsubara [20], based on specimens from Heta, Suruga, Japan. Morphological differences have also been found between Atlantic and Pacific specimens [21], which could in fact represent different species from different geographical areas, as inferred from Hulley and Paxton [2], but this aspect is still to be confirmed.

Therefore, all these clues seem to indicate the presence of a hidden biodiversity in the genus *Neoscopelus*, with cryptic species still waiting to be discovered. Hence, the specimen described here is preventively named as *N. cf. microchir*, pending a taxonomic revision of this genus.

Among the Myctophiformes, although the six rare neoscopelids show few morphological specializations, the divergent myctophids have evolved into about 250 species [25]. The large difference found in the number of species in the two families of the same order is due to the rate of diversification related to species-specific bioluminescence [26]. Myctophids, with lateral body photophores for species recognition, are diversifying into new species at a faster rate than neoscopelids, which have only ventral photophores of camouflage function that would not promote isolation of populations [26]. This theory would be contrary to finding greater diversity in neoscopelids. However, as a differential feature, the genus *Neoscopelus* is the only genus in its family to have photophores and, therefore, this characteristic could be related to greater diversity, in the order of several other fish genera also having ventral photophores such as *Cyclothone* Goode & Bean, 1883 (thirteen species), *Argyripnus* Gilbert & Cramer, 1897 (seven species), *Argyropelecus* Cocco, 1829 (seven species) or *Stomias* (Brauer, 1902) (nine species).

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