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First record of *Lepidonotus tenuisetosus* (Annelida: Polynoidae) from Tunisia with distributional notes

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Abstract

Four species of the genus *Lepidonotus* (Annelida, Polynoidae) have been listed in the Mediterranean, including *L. carinulatus* (Grube, 1869), *L. squamatus* (Linnaeus, 1758), *L. tenuisetosus* (Gravier, 1902) and *L. clava* (Montagu, 1808), although recent studies confirmed only the presence of the two latter. Based on materials collected in the Radès Area (Gulf of Tunis, Western Mediterranean), we provide a characterization of a population of *L. tenuisetosus* based on key morphological characters and body width/length relationships and conclude that all previously known Mediterranean reports from Israel, Croatia, Greece, Spain and Turkey did not belong to *L. tenuisetosus*, but either to *L. carinulatus* , *L. clava* or to other, non-identified species. Therefore, our finding represents not only the first correct record of *L. tenuisetosus* for the Tunisian waters and the Western basin, but for the whole Mediterranean Sea, and allows us to discuss on the possible reasons explaining the presence of this and other species of *Lepidonotus* in the Mediterranean.

Keywords: scale-worms; polychaetes; introduced species; new report; Gulf of Tunis; Mediterranean.

Introduction

Polynoidae is the second most species-rich polychaete family after Syllidae (Pamungkas *et al.*, 2019), currently including about 880 valid species (Read & Fauchald, 2022). Like all Aproditiformia, members of this family are also easily recognizable by the presence of dorsal elytra or scales, thus commonly known as scale-worms. Most of them appear to be carnivorous, whose diet comprises different invertebrates (including other polychaetes) (Plyuscheva *et al.*, 2010), while there is also evidence of deposit-feeding and macroalgal ingestion (Jumars *et al.*, 2015). In addition, they are moving and sit-and-wait predators (Jumars *et al.*, 2015). They are also among the families including the highest number of symbionts, representing more than 25% of the currently known species of Polynoidae, and about 45% of the currently known symbiotic polychaetes (Martin & Britayev, 1998, 2018). Polynoid scale-worms have been reported from virtually all oxic benthic marine environments (Jumars *et al.*, 2015), and only six species have been reported as introduced (Çinar, 2013). Three of them are Indo-Pacific/Red Sea species that entered the Mediterranean through the Suez Canal (i.e., Lessepsian migrants), of which two belong to *Lepidonotus* Leach, 1816 (Çinar, 2013), a genus containing about 80 valid species (Read & Fauchald, 2022) and occurring from intertidal to bathyal marine environments worldwide (De Assis *et al.*, 2015).

Among *Lepidonotus*, four species have been previously reported as being present in the Mediterranean, including *Lepidonotus carinulatus* (Grube, 1869), *Lepidonotus clava* (Montagu, 1808), *Lepidonotus squamatus* (Linnaeus, 1758) and *Lepidonotus tenuisetosus* (Gravier, 1902) (Barnich & Fiege, 2003), with only *L. clava* and *L. squamatus* being present in Tunisia (Zaâbi *et al.*, 2012). However, Barnich & Fiege (2003) only confirmed the presence of *L. tenuisetosus* and *L. clava*, with the former being considered as likely non-indigenous and the latter as native species for the Mediterranean Sea. *Lepidonotus tenuisetosus* was then reported in Turkey, Greece, and Croatia (Çinar, 2009; Mikac, 2015; Chatzigeorgiou *et al.*, 2016; Faulwetter *et al.*, 2017; Mikac *et al.*, 2020; Çinar *et al.*, 2021), based on the description of the species by Barnich & Fiege (2003) (M.E. Çinar, B. Mikac, G. Chatzigeorgiou, personal communications), and also in Spain, in a supplementary material annex listing the invertebrates from the bottoms surrounding mammal bones experimentally deployed off Blanes by Taboada *et al.* (2016).

In this paper, we: (1) fully describe and illustrate the

Tunisian specimens of *L. tenuisetosus*, (2) characterize the studied population based on the body width/length relationships, (3) compare them with previous descriptions of Mediterranean (Barnich & Fiege, 2003) and Gulf of Aden and Red Sea (Wehe, 2006) specimens, (4) examined all previous Mediterranean reports of *L. tenuisetosus*, and (5) discuss on the presence of the species of *Lepidonotus* in the Mediterranean, including their possible non-indigenous origin.

Material and Methods

The specimens of *L. tenuisetosus* were collected monthly from March 2018 to March 2019 in the Radès Area, Gulf of Tunis, (36.81°N, 10.28917°E) (Fig. 1A-C), among algae on a rocky bottom at 0-0.5 m depth (Fig. 1D). We used a 25x25 cm quadrat (Fig. 1D) to carefully scrape all inside surface contents, which were immediately placed in a hermetic plastic bag and fixed with 70% ethanol. In the laboratory, all polychaetes were sorted under a binocular stereomicroscope, identified to the lowest possible taxonomic level and preserved in 70% ethanol. The specimens of *L. tenuisetosus* were separated into individual vials for further morphological and morphometrical observations. Light microscopy photos were taken with a CMEX 5 digital camera connected to a Zeiss Stemi CS-2000-C stereomicroscope (body) and with a SP100 KAF1400 digital camera connected to a Zeiss Axioplan compound microscope (chaetae).

We estimated the relationships between body length and width. Among all specimens to characterize the Tunisian population, thirty were complete and were measured (in ventral view) under a binocular stereomicroscope (Zeiss Stemi 2000-C) using millimetric paper (body length, mm) and a calibrated micrometric ocular (body width with and without parapodia, at the level of chaetiger 5). The whole set of measurements is available from authors upon request. Linear regressions were calculated with the XLSTAT software, version 18.03.35937 (copyright Addinsoft 1995-2020), with their goodness-of-fit expressed by the corresponding coefficients of determination (R^2) and significance level (p), and then plotted using the appropriate routines of this software. The same

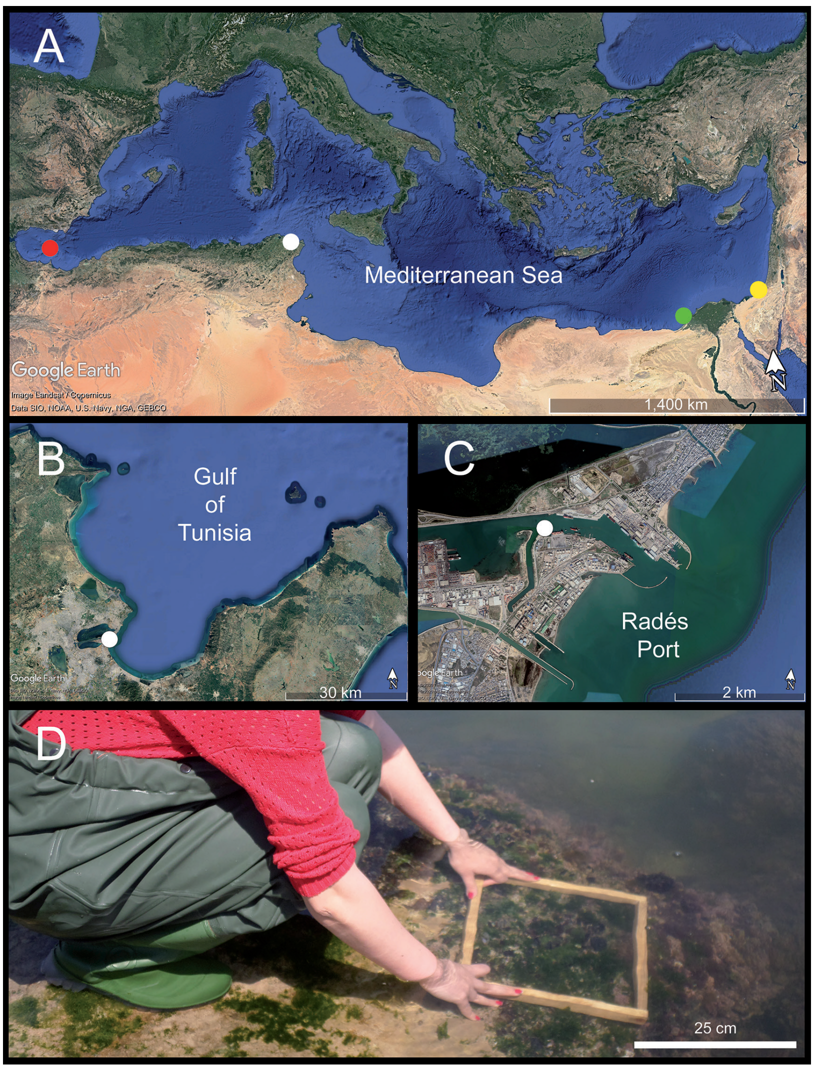

Fig. 1: A. Validated Mediterranean locations for *Lepidonotus tenuisetosus* (white spot: Gulf of Tunis) and *Lepidonotus carinulatus* (red spot: Alborán Sea; green spot: Peninsula of Sinai). B. Location of Radès area (white spot) in the Gulf of Tunis. D. Collecting site and method at Radès area. A–C: photos from Google Earth (images: © 2020 Landsat/Copernicus, TerraMetrics, Maxar Technologies; data: SIO, NOAA, U. S. Navy, NGA, GEBCO); D: photo by Marwa Chaibi.

software was used to run a correlation analysis to assess the strength of the width/length relationships (expressed by the Pearson correlation coefficient, P, and its corresponding *p*). All Tunisian specimens are deposited in the collections of the Centre d'Estudis Avançats de Blanes (CEAB).

The specimens from Croatia, Greece and Turkey attributed to *L. tenuisetosus* and not illustrated in the related references [Çinar, 2009 (only an elytron was drawn); Mikac, 2015; Chatzigeorgiou *et al.*, 2016; Faulwetter *et al.*, 2017; Mikac *et al.*, 2020; Çinar *et al.*, 2021] have been re-examined. The Natural History Museum of Rijeka (NHMR) gently loaned the Croatian specimen. Thus, it has been directly observed and photographed by the authors. The specimens from Greece, which are deposited at the Institute of Marine Biology, Biotechnology and Aquaculture of the Hellenic Centre for Marine Research (HCMR), were observed and photographed in a joint online session between the authors and Dr. G. Chatzigeorgiou, using a microscope with a digital camera linked to a computer and then sharing computer screens. The specimens from Turkey (deposited at the Museum of Ege University Faculty of Fisheries-ESFM), were examined by Dr. M. E. Çinar, who communicated his observations to the authors. The information on the specimens cited in Spain by Taboada *et al.* (2016) was requested to Dr. S. Taboada, who personally informed the authors about its status.

Results

A total of 265 specimens of *L. tenuisetosus* were collected during the study year. The maximum density was recorded in March 2018 (1472 individuals/m^2), followed by April 2018, August 2018, November 2018, and January-March 2019 (160-272 individuals/m^2). The lowest density (i.e., 80 individuals/m^2) was observed in May and October 2018.

Systematics

Family Polynoidae Kinberg, 1856
Subfamily Lepidonotinae Willey, 1902
Genus *Lepidonotus* Leach, 1816
Type species: *Aphrodita clava* Montagu, 1808

Lepidonotus tenuisetosus (Gravier, 1902)
Euphione tenuisetosa. Gravier (1902): 222-226, text-figs. 228-231, pl. 8 figs. 123-126.
Lepidonotus tenuisetosus. (Wehe, 2006); 107-109, fig. 24 a-l. Non Barnich & Fiege (2003): 86, fig. 44; non Çinar (2009): 2286, fig. 2A.

Material examined. CEAB.AP.985, 30 specimens, collected from Mars 2018 to Mars 2019 in Radès Area, Gulf of Tunisia (36.809424 N; 10.285653 E) by Marwa Chaibi, preserved in 70% ethanol.

Additional material. NHMR, PMR-17630, 1 specimen, preserved in 70% ethanol, Croatia, Adriatic Sea, 30/07/2007, collected by B. Mikac. HCMR_Nag_EL_01_2008_0185 and _0194, 2 specimens, preserved in 96% ethanol, 18/06/2008, rocky substrate with photophilic algae, (35.4233°N, 24.9838°E), Crete, collected by G. Chatzigeorgiou.

Description. Complete specimens with body 7.5-18.5 mm long, 1.36-2.44 wide (without parapodia), 2.08-4.08 mm wide (with parapodia) with 26 segments (25 chaetigers); elongated, flattened dorsoventrally (Fig. 2A-B), subrectangular in cross-section. Prostomium bilobed, without cephalic peaks; median antenna smooth, with ceratophore in anterior notch, fused to prostomium, style smooth, gradually tapering, without subterminal swelling; lateral antennae smooth, inserted terminally, with ceratophores fused to prostomium, styles smooth, tapering progressively, without subterminal swelling; palps smooth and gradually tapering (Fig. 2C-E). Two pairs of eyes close to each other, dorsolaterally on widest part of prostomium (Fig. 2C). Facial tubercle present. Pharynx with 18 big, conical, terminal papillae (Fig. 2D-F). Tentaculophores inserted laterally to prostomium, with few chaetae and a pair of dorsal and ventral tentacular cirri with smooth, gradually tapering styles. Second segment with first pair of elytra, sub-biramous parapodia and long, tapering, ventral cirri. Twelve pairs of elytra (Fig. 1A) on elytrophore segments (Fig. 3A) (2, 4, 5, 7, 9, 11, 13, 15, 17, 19, 21, 23), fully covering body dorsally; first pair round, second reniform (Fig. 2G), then progressively oval (Fig. 2H); with marginal fringing papillae, mostly digitiform but with some small, globular (Fig. 2I); surface with some central and posterior small macrotubercles with blunt or warty tips (Fig. 2I, 2J) and conical to cylindrical microtubercles (Fig. 2I-2K). Cirrophorous segments with dorsal cirri with smooth styles, gradually tapering, without subterminal swelling, reaching beyond tips of neurochaetae; styles of ventral cirri smooth, tapering, shorter than neuropodia; nephridial papillae well-visible, digitiform (Fig. 3B). Parapodia sub-biramous, noto- and neuropodia with elongate acicular lobe; tips of noto- and neuroacicula penetrating epidermis (Fig. 3A, 3B). Notochaetae slender, with numerous rows of small spines and tapering, capillary tips (Fig. 4A, 4B); neurochaetae of chaetiger 1 (i.e., second segment) slender than those of remaining chaetigers, with numerous rows of spines, similar to notochaetae but shorter (Fig. 3C); from chaetiger 3, all neurochaetae with unidentate tips, stouter than notochaetae, falcate, with numerous rows of spines in distal region (Fig. 3D). Nephridial papillae from segment 8 (chaetiger 7) onwards.

Remarks. The specimens from Tunisia fully agree with the original description by Gravier (1902), except for the special neurochaetae of chaetiger 1 (i.e., segment2), which do not show the terminal tuft of long, tight filiform extensions illustrated by Gravier (1902) in his fig. 231. Particularly, our specimens agree with the revision of types and additional materials by Wehe (2006), (Tables 1 and 2, Fig. 5A-5E), including the shape of the neurochaetae of chaetiger 1, which he described as "*Neurochaetae of second segment altered (Fig. 24 l),*

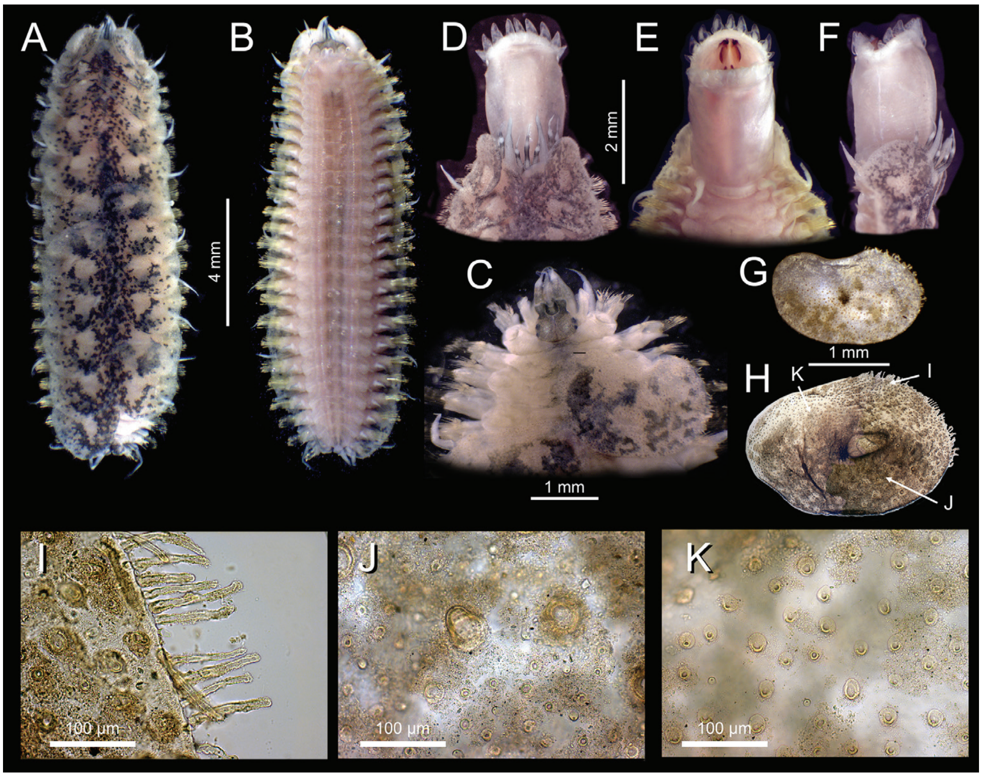

Fig. 2: *Lepidonotus tenuisetosus*. A. Whole body, dorsal view. B. Whole body, ventral view. C. Anterior region (without elytra), dorsal view. D. Anterior region (pharynx everted), dorsal view. E. Anterior region (pharynx everted), ventral view. F. Anterior region (pharynx everted), lateral view. G. Second elytra. H. Tenth elytron. I. Detail of fringing papillae from the tenth elytron. J. Detail of macrotubercles from the tenth elytron. K. Detail of microtubercles from tenth elytron.

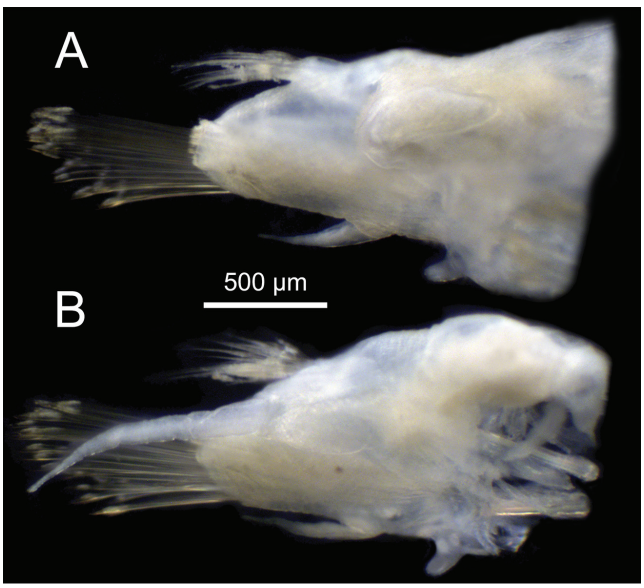

Fig. 3: *Lepidonotus tenuisetosus*. A. Elytrophorous parapodium. B. Cirrophorous parapodium.

more slender, with more numerous rows of spines and fine tips" and illustrated in his figure 24 l as lacking the terminal tuft of filiform extensions (Wehe, 2006). Conversely, they differ from the Sinai specimens reported by Barnich & Fiege (2003), specifically in the shape of the antennae and dorsal cirri, in the eye position, the types of micro- and macrotubercles and the shape of neurochaetae tips (Fig. 5F-5I), which fully agree with the features of *L. carinulatus* (Tables 1 and 2, Fig. 5J-Q).

The very small specimen (i.e., an anterior fragment with only eight chaetigers, measuring about 1.6 mm long and 0.4 mm wide) reported as *L. tenuisetosus* from the Croatian coasts of the northern Adriatic Sea is in very poor conditions, lacking elytra and all appendages (Fig. 6A, 6B). However, the chaetae are well-visible, being uni-and bidentate, with several rows of spines and the distal ones much longer than the basal ones (Fig. 6C-6E). Therefore, if it belongs to *Lepidonotus* (which cannot be confirmed considering its present conditions), it could be *L. carinulatus*, but certainly not *L. tenuisetosus*.

The specimens from Crete show unidentate chaetae, but the antennae and cirri have distal swellings and

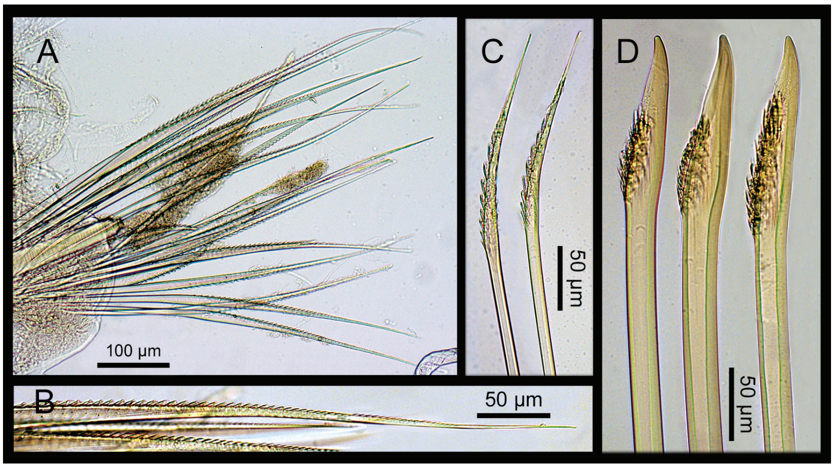

Fig. 4: *Lepidonotus tenuisetosus*. A. Notochaetae from chaetiger 15. B. Tip of notochaeta. C. Neurochaetae from first chaetiger. D. Neurochaetae from chaetiger 15.

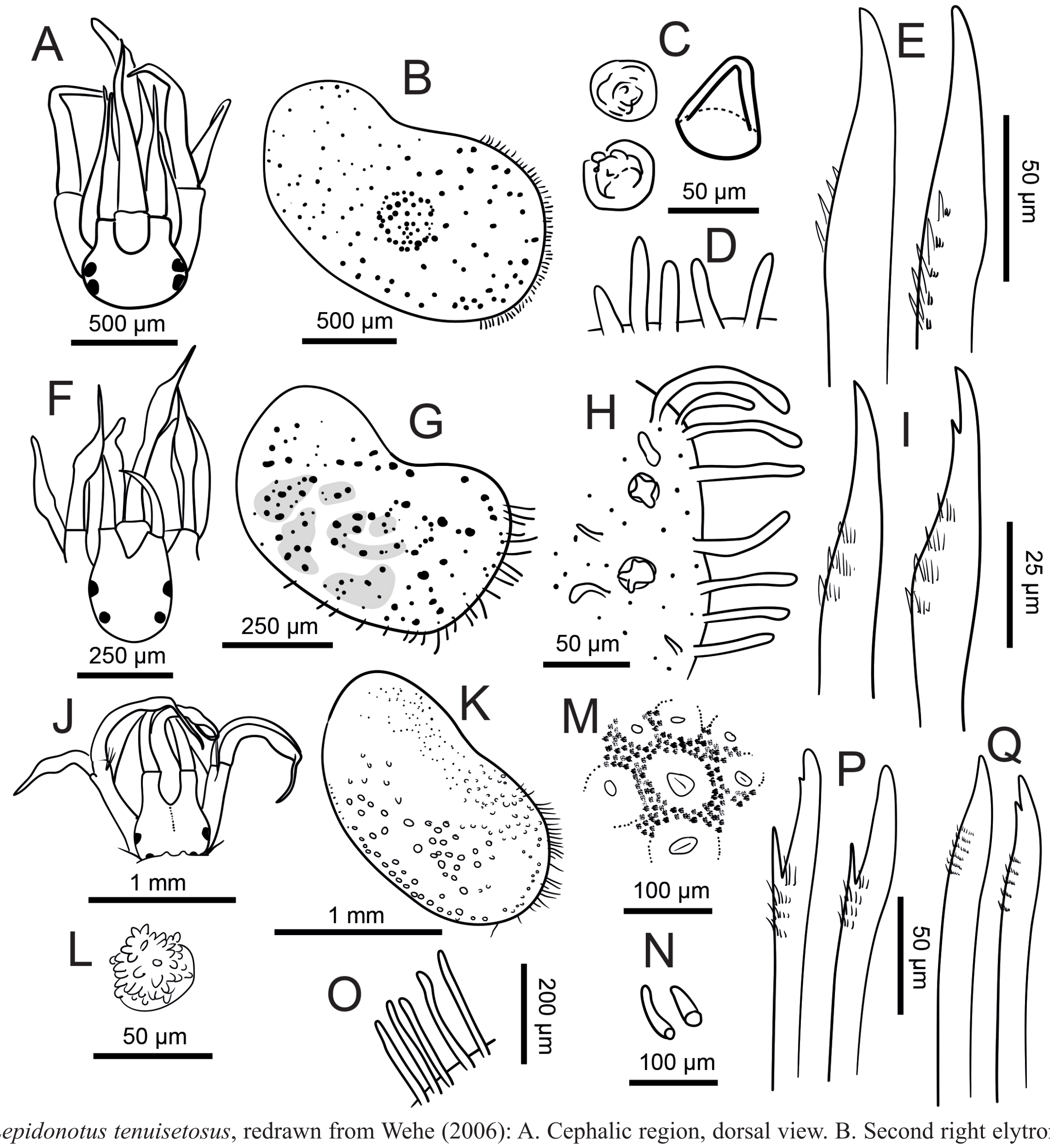

Fig. 5: *Lepidonotus tenuisetosus*, redrawn from Wehe (2006): A. Cephalic region, dorsal view. B. Second right elytron. C. Macrotubercles. D. Fringing papillae. E. Neurochaetae. *Lepidonotus carinulatus*, redrawn from Barnich & Fiege (2003): F. Cephalic region, dorsal view. G. Third right elytron. H. Lateral margin showing macrotubercles, digitiform papillae and fringing papillae. I. Neurochaetae. *Lepidonotus carinulatus*, redrawn from Wehe (2006): J. Cephalic region, dorsal view. K. Second right elytron. L. Macrotubercle. M. Carinate microtubercles and pigmentation. N. Digitiform papillae. O. Fringing papillae. P. Upper neurochaetae. Q. Lower neurochaetae.

Table 1. Main characters distinguishing between *Lepidonotus tenuisetosus sensu* Barnich & Fiege (2003) and Wehe (2006) and *Lepidonotus carinulatus sensu* Wehe (2006).

	Lepidonotus tenuisetosus		*Lepidonotus carinulatus*
Characters	**Barnich & Fiege (2003)**	**Wehe (2006)**	**Wehe (2006)**
Median ceratophore	Slightly inflated subdistally	Tapering to terminal filum, no subterminal swelling	Slightly inflated subdistally
Eye pairs on prostomium	Anterior dorsolateral on widest part, posterior dorsally near hind margin	Close to each other, dorsolateral	Anterior dorsolateral on widest part, posterior dorsally near hind margin
Dorsal cirri	Slightly inflated subdistally	Tapering to terminal filum, no subterminal swelling	Slight subterminal swelling and terminal filum
Elytra microtubercles	Cylindrical with multifid, crown-like tips	Conical to globular	Posterolateral warty, with nodules or pointed projections
Elytra macrotubercles	Absent	Present centrally and posteriorly with blunt or warty tips	Centrally conical and globular, posterolateral warty
Neurochaetae 1st chaetiger	Non-defined	Slender, with numerous rows of spines and fine tips	Slender, with numerous rows of spines and fine tips
Other neucochaetae	Mostly unidentate, a few minutely bidentate; with numerous rows of spines	All unidentate, with few rows of spines	Bidentate, with small secondary tooth, lower ones apparently unidentate; with several rows of spines, distal ones usually much larger than basal

Table 2. Dichotomous key showing the main characters allowing distinguishing the species of *Lepidonotus* mentioned in this paper. *: Specimens directly observed by the authors.

	Characters		Species	Location	References
1a	Elytra with smooth margins	——	*Lepidonotus clava*	Greece*	Faulwetter *et al.* (2017); Chatzigeorgiou *et al.* (2016)
				Mediterranean Sea	Barnich & Fiege (2003)
1b	Elytra with fringing papillae	— 2			
2a	Neurochaetae uni- and bidentate; antennae and dorsal cirri slightly inflated subdistally	——	*Lepidonotus carinulatus*	Sinai	Barnich & Fiege (2003)
				Alboran Sea	Baratech *et al.* (1986)
				Red Sea, Arabian Sea, Indo-West Pacific	Wehe (2006)
2b	Neurochaetae unidentate; antennae and dorsal cirri tapering to terminal filum	— 3			
3a	Eyes dorsoventrally on prostomium; microtubercles with blunt or warty tips	——	*Lepidonotus tenuisetosus*	Gulf of Aden	Gravier (1902)
				Red Sea	Wehe (2006)
				Arabian Sea	Wehe (2006)
				Tunisia *	This paper
3b	Anterior pair of eyes dorsolaterally on widest part, posterior dorsally near hind margin; filiform papillae on elytra	——	*Lepidonotus* sp.	Turkey	Çinar (2009, 2013)

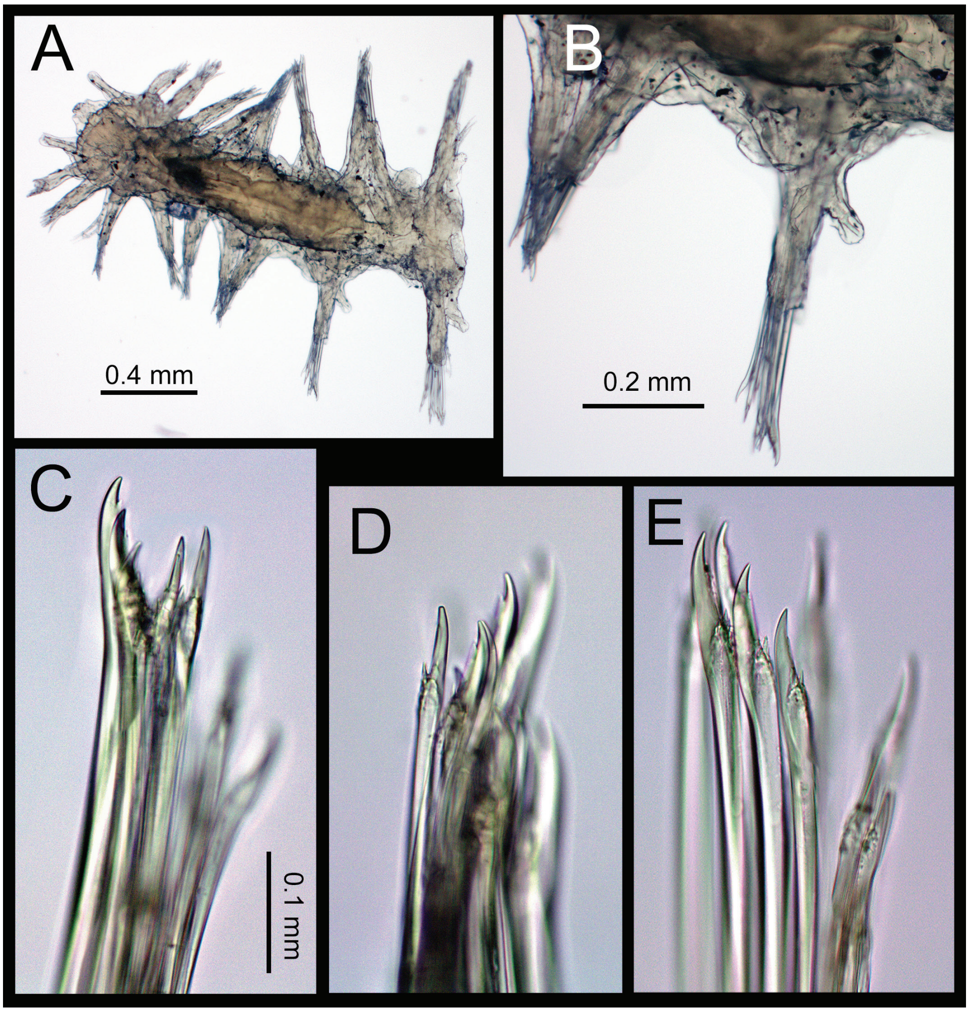

Fig. 6: *Lepidonotus* sp., NHMR PMR-17630, Croatia: A. Anterior end, dorsal view. B. Parapodia 7 and 8, right side. C. Neurochaetae from parapodium 8 (right side). D. Neurochaetae from parapodium 8 (left side). E. Neurochaetae from parapodium 9 (right side).

pigmented bands and, most importantly, the elytra have smooth margins (Fig. 7A-7E). Therefore, they belong to *L. clava*.

Upon the authors' request, M. E. Çinar re-examined the specimens from Mersin Bay in Turkey–ESFM-POL/05-547 (n=17, 18/09/2005, K15, 0.2=3 m, stones with algae, approx. (36.7167°N, 34.5°E). Mersin Bay) and ESFM-POL/2005-547 (n=1, 19/09/2005, K17, 0.1-2 m, stones with algae, approx. 36.4833°N, 34.1833°E). Although all specimens show unidentate chaetae with all spines of a similar size, the eyes are not in dorsolateral position and the elytra show filiform papillae, which allow us confirming them as not belonging to *L. tenuisetosus*, being likely a different introduced or undescribed species that will require further analyses to be defined (M.E. Çinar, personal communication).

Finally, the specimens included in the faunal list of the invertebrates from the bottoms surrounding mammal bones experimentally deployed in Blanes Bay (Appendix S1 in Taboada *et al.*, 2016) are probably lost and their identification was just tentative (S. Taboada, personal communications). Therefore, its real identity cannot be currently determined and further sampling is required to confirm which species is present in Blanes Bay. However, it must be taken into account that previous studies only reported *L. clava* (e.g., Camp, 1976; Campoy & Jordana, 1978; Martin, 1987; Plyuscheva & Martin, 2009).

Distribution. Gulf of Aden, Red Sea, and Arabian Sea (Gravier, 1902; Wehe, 2006); Gulf of Tunisia, Mediterranean Sea (present study). Doubtful records: India, Vietnam, South China Sea, and New Caledonia (Uschakov, 1982; Hanley, 1992; Barnich *et al.*, 2004; Wehe, 2006). Previous records from Croatia (Mikac, 2015; Mikac *et al.*, 2020), Greece (Chatzigeorgiou *et al.*, 2016; Faulwetter *et al.*, 2017), Turkey (Çinar, 2009; Çinar *et al.*, 2021), and Israel (Barnich & Fiege, 2003; Zenetos *et al.*, 2010; Çinar, 2013) are discarded according to our results.

Habitat. Common on rocks (Red Sea); found on a rocky bottom among algae (Tunisia). 0-0.5 m depth.

Morphometry

Body width without parapodia (WoP) ranged from 0.38 to 2.76 mm (1.90 ± 0.48 mm on average) and with parapodia (WP), from 0.53 to 3.45 (3.10 ± 0.86 mm on average). Body length (L) ranged from 2.23 to 18.50 mm (12.74 ± 3.73 mm on average). Both WoP and WP

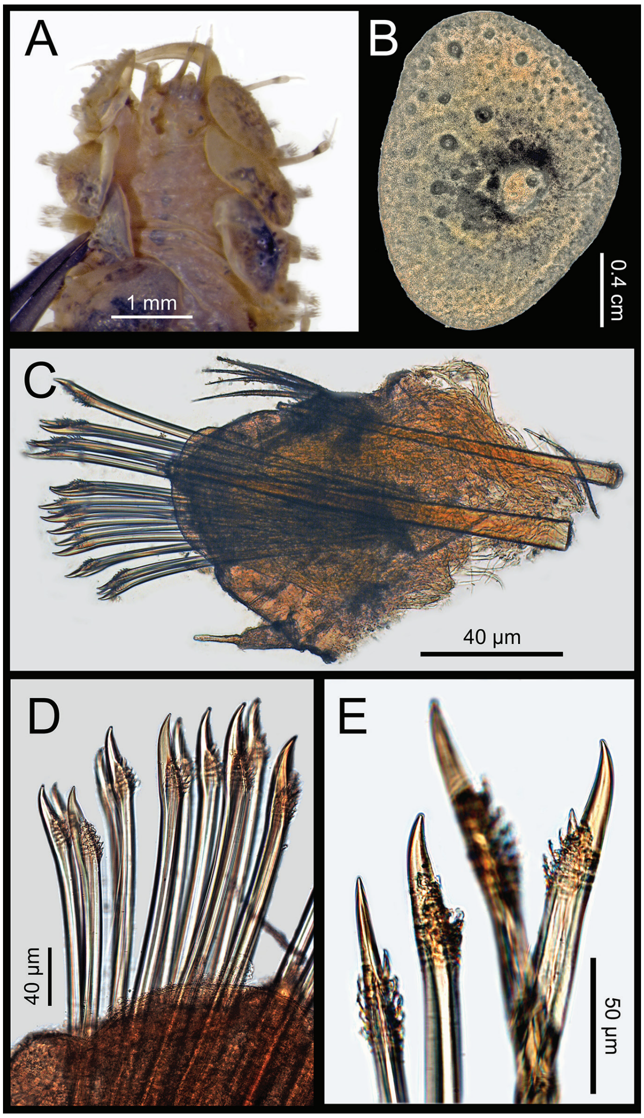

Fig. 7: *Lepidonotus clava* from Greece. HCMR_Nag_EL_01_2008_0185. A. Anterior end, dorsal view. B. Midbody elytron. C. Midbody elytrophorous parapodium. D. Midbody neurochaetae. HCMR_Nag_EL_01_2008_0194: E. Detail of midbody neurocheatae tips. Photos by G. Chatzigeorgiou.

showed highly significant positive relationships with L (Fig. 8A, 8B). Based on the studied population, the WP vs. L relationship explained a slightly higher percentage of the observed variability (ca. 70%) than that of WoP (ca. 66%). Thus, WP seems to be an adequate descriptor for further monitoring *L. tenuisetosus*, although this must be validated by acquiring additional data from different populations of the species.

Discussion

Lepidonotus was established by Leach (1816) without providing a generic diagnosis. Much later, the Lepidonotinae were extensively reviewed, including descriptions and keys for more than 50 species of *Lepidonotus* (Seidler, 1923). This review lacked illustrations for most of these species and did not clearly state whether type specimens were examined. However, it provided excellent baseline information on most genera of Lepidonotinae (Wehe, 2006). A more recent analysis of the Mediterranean Aphroditoidea concluded that only *L. clava* and *L. tenuisetosus* occurred in Mediterranean waters, while *L. squamatus* appeared to be limited to East Atlantic coasts and *L. carinulatus* seemed to be an Indo-Pacific/Red Sea species (Barnich & Fiege, 2003).

Lepidonotus clava is the most typical Mediterranean species of the genus (Barnich & Fiege, 2003). It can be distinguished from *L. tenuisetosus* by lacking fringing marginal papillae on elytra (present in *L. tenuisetosus*) and having rugose micro- and macrotubercles, the latter very numerous (cylindrical microtubercles and few blunt or warty macrotubercles in *L. tenuisetosus*). *Lepidonotus tenuisetosus*, together with *L. squamatus* and *L. carinulatus,* bears elytra with marginal papillae, but differs in having slender notochaetae with capillary tips and neurochaetae with unidentate tips only (exclusively bidentate in *L. squamatus*, unidentate and a few minutely bidentate in *L. carinulatus*), as well as cylindrical microtubercles and a few blunt or warty macrotubercles (microtubercles with pointed projections, macrotubercles conical, globular, and warty in *L. carinulatus*).

Previous Mediterranean records of *L. squamatus* (e.g., Fauvel, 1937; Bellan, 1964; Pozar-Domac, 1978) have not been further confirmed, being tentatively attributed either to *L. clava* or, as the former has fringing papillae on elytra, more likely to *L. carinulatus* or *L. tenuisetosus*, which also have papillae on elytral margins (Barnich & Fiege, 2003). In the Mediterranean, the presence of *L. carinulatus* is currently known only from four specimens found in the Alborán Sea (Baratech *et al.*, 1986; Templado *et al.*, 1986) (Fig. 1A). Barnich & Fiege (2003) were not able to find any specimen of this species among the extensive materials they revised. Thus, they assumed that the Alborán specimens could have been introduced together with "pearl oysters". However, pearl oysters did not exist in the Alborán Sea.They were only mentioned by Baratech *et al.* (1986) as part of the summary of habitats of *L. carinulatus* extracted from the bibliography. By the 1980s, Alborán waters only harbored native common oysters (i.e., *Ostrea edulis* Linnaeus, 1758), with and the only attempts (unsuccessful) to cultivate them in the region dating from the beginning of the 21th century (Robles, 2010). This, together with the fact that the Alborán specimens were collected in coralligenous bottoms at 70-200 m depth, allows us to discard the hypothesis of an introduction through oyster culture. Alternatively, the species could be introduced another way, confused with a similar species, or native.

Lepidonotus carinulatus was originally described as *Polynoe (Lepidonotus) carinulata* (Grube, 1869) and the type locality was identified as "Rothes Meer" in the catalog of the Zoologischen Museum of Berlin. C.G. Ehrenberg and W. Hemprich collected type material, during the 1820-1825 expedition of the Berlin Academy to Libya, Egypt and both shores of the Red Sea, which also visited the Eastern Mediterranean (Alexandria, Egypt). The expedition was an organizational disaster, from which Ehrenberg was the only survivor. When Ehrenberg final-

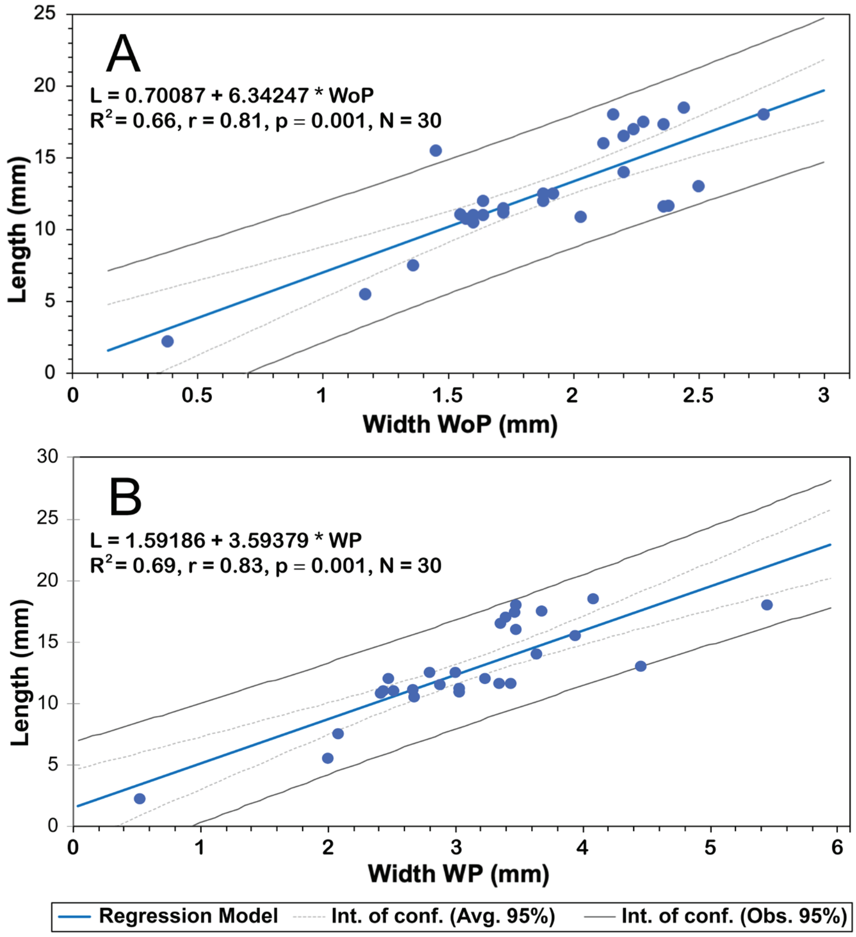

Fig. 8: Size relationships in *Lepidonotus tenuisetosus*. A. Body width without parapodia (WoP) *vs*. body length. B. Body width with parapodia (WP) *vs*. body length.

ly arrived in Berlin and returned to work on his material, he found that "...the remaining specimens were no longer to be found with their original, so exceptionally careful, labeling. On the contrary, this had been lost or mixed up, and locality and other important data could not be ascertained" (Heinstein 1877 in Baker 1997). Many specimens had either been sold as duplicates, exchanged, or given away. Moreover, the original labels (containing detailed data on collection localities and dates) were arbitrarily discarded and replaced by less informative ones. This crucial issue gave rise to an unsolvable situation, as some of Ehrenberg's materials were collected in Alexandria and not in the Red Sea (Grube, 1869: p. 485). "*Polynoe (Lepidonotus) carinulata*" was one of the species that lacked the original labels (Grube, 1869). Later, it was reported as being present in the Sea of Bohol (Philippines), with the Red Sea being assumed as the type locality (Grube, 1878). However, this was just an assumption, as the problem with the original labels described by Ehrenberg did not allow choosing between a possible origin in the Red Sea or the Eastern Mediterranean. Moreover, the Mediterranean specimens from Ehrenberg's expedition were collected in Alexandria before the opening of the Suez Canal in 1869. Thus, we cannot discard *L. carinulatus* as a Mediterranean native species, which could be eventually supported by its more recent record in the Western Mediterranean by Baratech *et al.* (1986).

Lepidonotus tenuisetosus was considered a non-indigenous species (NIS) in the Mediterranean. Apparently originating in the Indo-Pacific/Red Sea, it seemed to be a Lessepsian migrant, whose presence was only initially known from the coasts of the Peninsula of Sinai (Fig. 1A) (Barnich & Fiege, 2003). Three years later, Wehe (2006) provided detailed descriptions of *L. tenuisetosus* and *L. carinulatus* based on type specimens and numerous newly collected Arabian Sea materials. This allowed us to identify some inconsistencies in the description of the Eastern Mediterranean specimens of Barnich & Fiege (2003). For example, in *L. tenuisetosus*, antennae and cirri lack distal swelling, eye pairs are dorsolateral and close to each other, a peculiar position only found in a few species of *Lepidonotus* (Wehe, 2006), and neurochaetae have all unidentate tips (Table 1). In turn, Barnich & Fiege's (2003) Eastern Mediterranean specimens showed antennae and cirri with distal swelling, posterior eye pair placed dorsally near the hind of prostomial margin, and neurochaetae with both uni- and bidentate tips (Table 1). Moreover, a presumed "characteristic" trait, the elytral macrotubercles, may also be confusing, at least in *L. carinulatus*. A high inter-individual variability would lead two specimens at the two extremes of the range to be considered as different species. Only by comparing numerous individuals is it possible to perceive the existence of a gradual transition among specimens (Wehe, 2006). Thus, we conclude that the Barnich & Fiege's (2003) specimens belong to *L. carinulatus*, which would indirectly support Ehrenberg's 19th century specimens as being collected in Alexandria.

Further mentions of *L. tenuisetosus* as NIS in the Mediterranean (e.g., Zenetos *et al.*, 2010; Çinar, 2013; Núñez *et al.*, 2015) refer to the Israeli specimens mentioned by Barnich & Fiege (2003). Moreover, its reports from the coastal areas of Croatia, Greece and Turkey (Çinar, 2009; Mikac, 2015; Chatzigeorgiou *et al.*, 2016; Faulwetter *et al.*, 2017; Mikac *et al.*, 2020; Çinar *et al.*, 2021) have been checked for this study and discarded as belonging to *L. tenuisetosus*, while that in the north-eastern Iberian Peninsula by Taboada *et al.* (2016) cannot currently be supported. Therefore, our findings not only prove the presence of *L. tenuisetosus* in Tunisian waters, being the first report for the country, but also represent the first report of the species for the Western Mediterranean and actually for the whole Mediterranean Sea.

In light of the present data, we cannot entirely discard *L. tenuisetosus* as a native Mediterranean species, either previously overlooked or confused with other species of *Lepidonotus*. However, it seems more reliable to consider it a NIS, whose presence would constitute another example of the Mediterranean tropicalization process (Bianchi & Morri, 2003). It must also be pointed out that our study site at the Radès Port area (Fig. 1C) suffers from the influence of a heavy vessel traffic associated with the petrol industry (Bouhedi *et al.*, 2021). Thus, the possibility of the presence of *L. tenuisetosus* resulting from an introduction (e.g., through ballast waters) seems feasible. The extremely high density (i.e., from 80 to 1400 individuals/m^2) in our studied station is also remarkable, as the species of the genus habitually tend to be solitary or much less abundant. For example, only four specimens of *L. carinulatus* were found in Alborán in the eight stations studied, which corresponds to a very low density taking into account the enormous sample size collected by the "Italian bar" trawl, a gear used to sample red coral at that time (Baratech *et al.*, 1986). Indeed, we cannot assess whether the high density of *L. tenuisetosus* in spring could result from a recruitment event, or whether the observations made in our sampling station can be generalized to the whole Radès area. However, the overall high densities observed seem at least atypical and, most probably, not long-lasting. Instead, they strongly suggest an unstable situation resulting from opportunistic colonization, which may also be compatible with a relatively recent introduction.

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