Global Ecology and Conservation 32 (2021) e01920

Contents lists available at ScienceDirect

Global Ecology and Conservation

journal homepage: www.elsevier.com/locate/gecco

Annelid invaders in the Black Sea region: The distribution of *Streblospio gynobranchiata* and first occurrence of *Laonome xeprovala*

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ARTICLE INFO

Keywords:
Polychaeta
Black Sea
New records
Invasive species
Spatial distribution

ABSTRACT

The present study reports the spread of *Streblospio gynobranchiata* (Annelida, Spionidae) off Romanian coast and adjacent waters and the first occurrence of *Laonome xeprovala* (Annelida, Sabellidae) in the Black Sea basin. The spionid is considered among worst invaders in Mediterranean soft bottoms, while the sabellid is a new invader that successfully settled in the Baltic and the Azov Seas and, after a decade since first European report, finally reached the Black Sea (Musura Bay and Sakhalin Lagoon, Danube Delta). Based on 50 samples collected between 2011 and 2020, we provide an overview of the distribution and possible arrival pathways of both species. Their larval ecology and life traits suggest a secondary introduction through shipping. Since first report along the Romanian coast in 2011, *S. gynobranchiata,* has spread very quickly, reaching very high densities (4945.2 ± 7599.4 ind.m^{-2}) at 11–15 m depth in infralittoral sandy-mud habitat of the Danube plume area, while *L. xeprovala* remains confined to estuarine habitats, reaching its highest abundance (100.8 ind.m^{-2}) in Sakhalin Lagoon.

1. Introduction

Enclosed or semi - enclosed basins, such as the Black Sea, seem particularly sensitive to biological invasions (Leppäkoski et al., 2009). Many non-indigenous species (NIS) integrate into local ecosystems without significantly changing anything, but sometimes spread rapidly, causing economic, environmental human health harming (Zaitsev and Mamaev, 1997; Gomoiu et al., 2002; Zaitsev and Ozturk, 2001; Paavola et al., 2005; Galil et al., 2014; Alexandrov, 2015). Maritime transport and port activities are a long-lasting source of NIS, with ballast water as one of main vectors (Carlton, 1985; Grigorovich et al., 2003; Gollasch et al., 2007; Simkanin et al., 2009) while, more recently, climate change seems to be one of the main invasion drivers (Bellard et al., 2013, 2018). The increasing number of NIS in the Black Sea since 2000 seems to be related to climate change as it appears to be related to a parallel an increase in water temperature (Shalovenkov, 2020).

Black Sea is a particular marine basin, with special hydrological characteristics. Situated in the temperate Ponto-Caspian region, it has a long period of common ancient history with the Caspian Sea. Marine water enters via the Bosporus Strait while major rivers like Danube, Dnieper and Dniester contribute with the freshwater input. Its low salinity and low species diversity, combined with a high levels of eutrophication and trade rate, have facilitated the establishment of NIS with high ecological plasticity (Leppäkoski and

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https://doi.org/10.1016/j.gecco.2021.e01920
Received 8 July 2021; Received in revised form 8 November 2021; Accepted 9 November 2021
Available online 10 November 2021
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Mihnea, 1996), which have generated very acute problems. Often is difficult to estimate the pathways and the exact moment of NIS arrival, except when intentional. Frequently, they are recorded too late, when spatial-temporal viable populations have already been established.

As NIS introduction vectors are expected to further expand and diversify (Seebens et al., 2018), to provide accurate data on biological invasions it is important to find new and/or more efficient ways to prevent new invasions, as well as to minimize the damage caused by already established invasive NIS.

Some NIS have been implicated in the decline of marine ecosystems services (Harris and Tyrrell, 2001), such as *Ficopomatus enigmaticus* (Read and Gordon, 1991), *Hydroides elegans* and *H. dianthus* (Hirata and Akashige, 2004). In the Black Sea, the gastropod *Rapana venosa* feeds on bivalves and is responsible for the depletion of large stocks of commercial bivalves (*Mytilus galloprovincialis*) and the associated communities since the 1950 s (Zolotarev, 1996; Salomidi et al., 2012). On the other hand, some of these NIS are fished or farmed and have substantial positive impact on food provision, like *R. venosa* has supported very profitable fisheries in the Black Sea (Sahin et al., 2009).

To date, no major economic impact of invasive polychaetes has been observed on the Black Sea ecosystem despite there are 18 polychaetes (Table 1) among the 160 NIS have been identified since the first scientifically documented invasions of the barnacles *Amphibalanus improvisus* Darwin, 1854 and *A. eburneus* Gould, 1841 (Alexandrov, 2004; Leppäkoski et al., 2009). Of the 18 species (Table 1) (Gomoiu et al., 2002; Surugiu, 2008; Kurt-Şahin and Çinar, 2012; Boltacheva et al., 2021), some have an uncertain status (Kurt-Şahin and Çinar, 2012; Vinogradov et al., 2018).

NIS identification is a key point of the Marine Strategy Framework Directive (MSFD) (EC, 2008) and of the IAS Regulation (EU Regulation 1143/2014) that fulfils the Target 5 of the EU 2020 Biodiversity Strategy. However the implementation of preventing NIS spread requires reliable data on the distribution, pathways, and impacts for each species, which still lacks in most cases.

The polychaetes are usually a major component of marine benthos, and their study is crucial to understand ecosystem structure and functioning (Surugiu, 2005; Begun et al., 2018; Teaca et al., 2019, 2020). Moreover, polychaetes are frequently used as indicators of marine pollution, as the affectation to benthos well-being tends to be reflected in the variations of the structure of their communities (Dean, 2008). However, to be able to perform an accurate assessment of changes in ecosystems and benthic biodiversity critically depends on having proper identifications at, if possible, species level. To date, 238 polychaete species have been reported from the Black Sea (Kurt Şahin and Çınar, 2012). Out of them, 61 occur in Romanian soft bottom from 10 to 150 m depth (Teaca et al., 2021), with only eight being recognized as NIS and four having an invasive character (Table 1).

The aim of this paper is to contribute to the growing knowledge on NIS in the Black Sea and the Danube Delta lagoon systems by (1) presenting an overview of the expansion of invasive spionid *Streblospio gynobranchiata* (Spionidae Grube, 1850) along the Romanian Black Sea Coast since first record of it in 2011 and till 2020 and (2) reporting on the presence of a new NIS, the polychaete *Laonome xeprovala* (Sabellidae Latreille, 1825) based on material collected in 2018.

2. Materials and methods

2.1. Description of the area

The Romanian coastline is divided into two units, Northern (between Musura Bay and Cape Midia Harbour jetties) and Southern

Table 1
List of alien polychaetes species established in the Black Sea during the 20th century.

No.	Species	Reference	
		Black Sea	Romania
1	****Ficopomatus enigmaticus** (Fauvel, 1923)	Annenkova (1929)	Dumitrescu (1962)
2	***Hesionides arenaria** Friedrich, 1937	Valkanov (1954)	Surugiu (2006)
3	***Streblospio shrubsolii** (Buchanan, 1890)	Marinov (1957)	Manoleli (1980)
4	***Magelona mirabilis** (Johnston, 1865)	Marinov (1959)	Dumitrescu (1963)
5	***Nephtys ciliata** (Müller, 1788)	Rullier (1963)	
6	***Capitellethus dispar** (Ehlers, 1907)	Rullier (1963)	
7	***Sigambra tentaculata** (Treadwell, 1941)	Kiseleva (1964)	
8	****Polydora cornuta** Bosc, 1802	Losovskaya and Nesterova (1964); Radashevsky and Selifonova (2013)	Surugiu (2005)
9	***Streptosyllis varians** Webster & Benedict, 1887	Kaneva-Abadzieva and Marinov (1966); Marinov (1966)	
10	*Glycera capitata* Örsted, 1843	Morduhai Boltovskoy (1972)	
11	***Polydora websteri** Hartman in Loosanoff & Engle, 1943	Surugiu (2005)	Surugiu (2005)
12	****Dipolydora quadrilobata** (Jacobi, 1883)	Todorova and Panayotova (2006)	Surugiu (2009)
13	****Streblospio gynobranchiata**Rice and Levin (1998)	Boltacheva (2008); Radashevsky and Selifonova (2013)	Teaca et al. (2015)
14	*Hydroides dianthus* (Verrill, 1873)	Boltacheva et al. (2011)	
15	*Prionospio pulchra* Imajima, 1990	Dağli et al., 2011	
16	*Pseudopotamilla* cf. *reniformis* (Bruguière, 1789)	Boltacheva and Lisitskaya (2016)	
17	*Marenzelleria neglecta* Sikorski & Bick, 2004	Syomin et al. (2017)	
18	*Hyboscolex* cf. *pacificus* (Moore, 1909)	Lisitskaya et al. (2019)	

Note: *- species with an uncertain invasive status.
** - species with invasive character in the Romanian Black Sea coast.

(up to Vama Veche), the later including the most diverse geomorphological features (Panin and Jipa, 1998; Panin, 2005). The highly dynamic longshore southward drift of Danube River sediments characterizes the Northern unit (Panin and Jipa, 1998), together with massive freshwater inflows and 75% of total nutrient input to the Black Sea (Mee, 1992) that came not only from the Danube but also from Ukrainian seaside (Cociasu and Popa, 2004). The entire NW Black Sea sector receives 80% of the basin's total freshwater input, originating from the Danube (208 km^3 yr^{-1}), Dnieper (43.4 km^3 yr^{-1}) and Dniester (9.1 km^3 yr^{-1}) rivers (Mikhailov and Mikhailova, 2008).

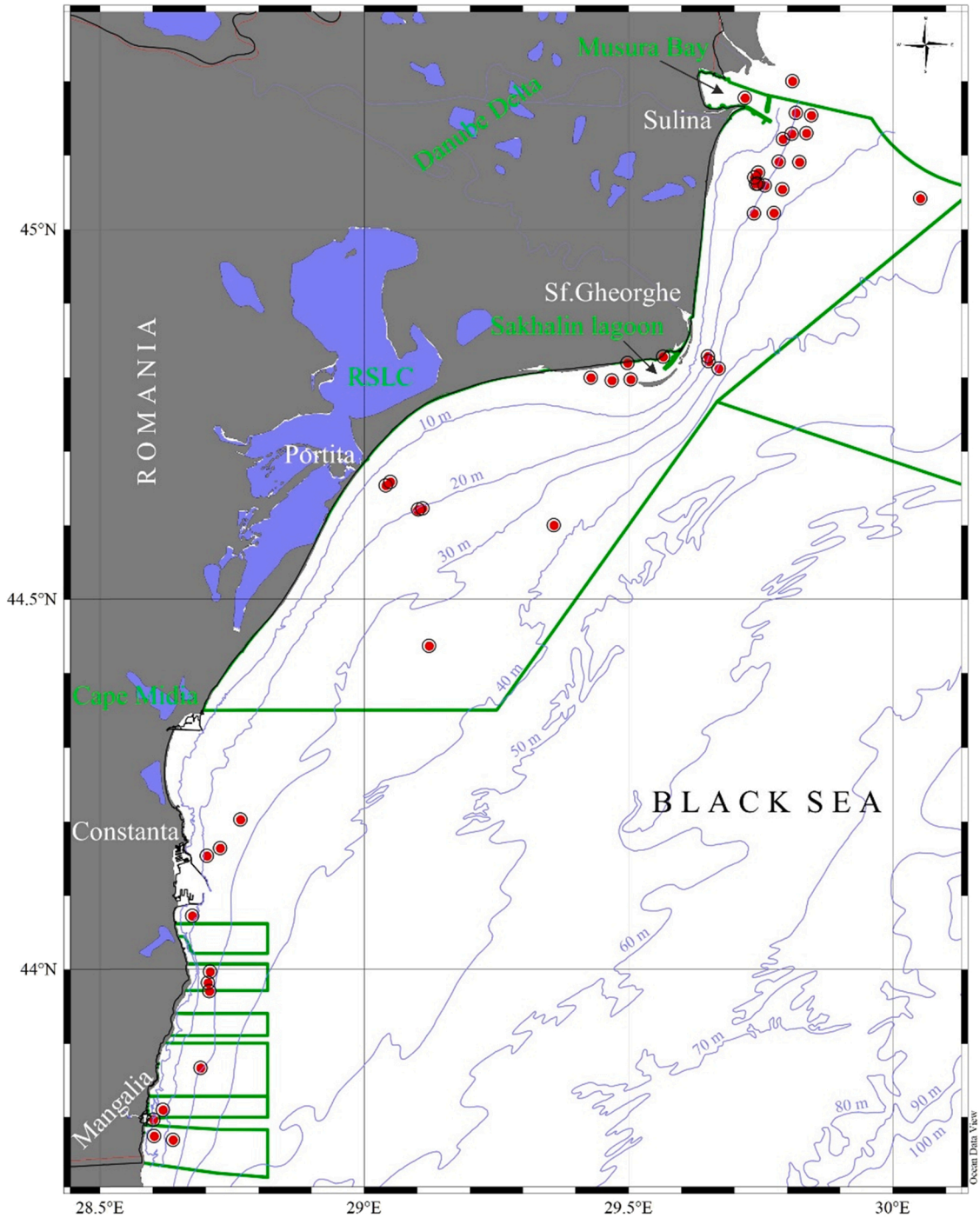

Fig. 1. Sampling sites locations where *S. gynobranchiata* and *L.xeprovala* were recorded (time period: 2011–2020) of the Romanian Black Sea coast (green polygons: MPA`s). (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

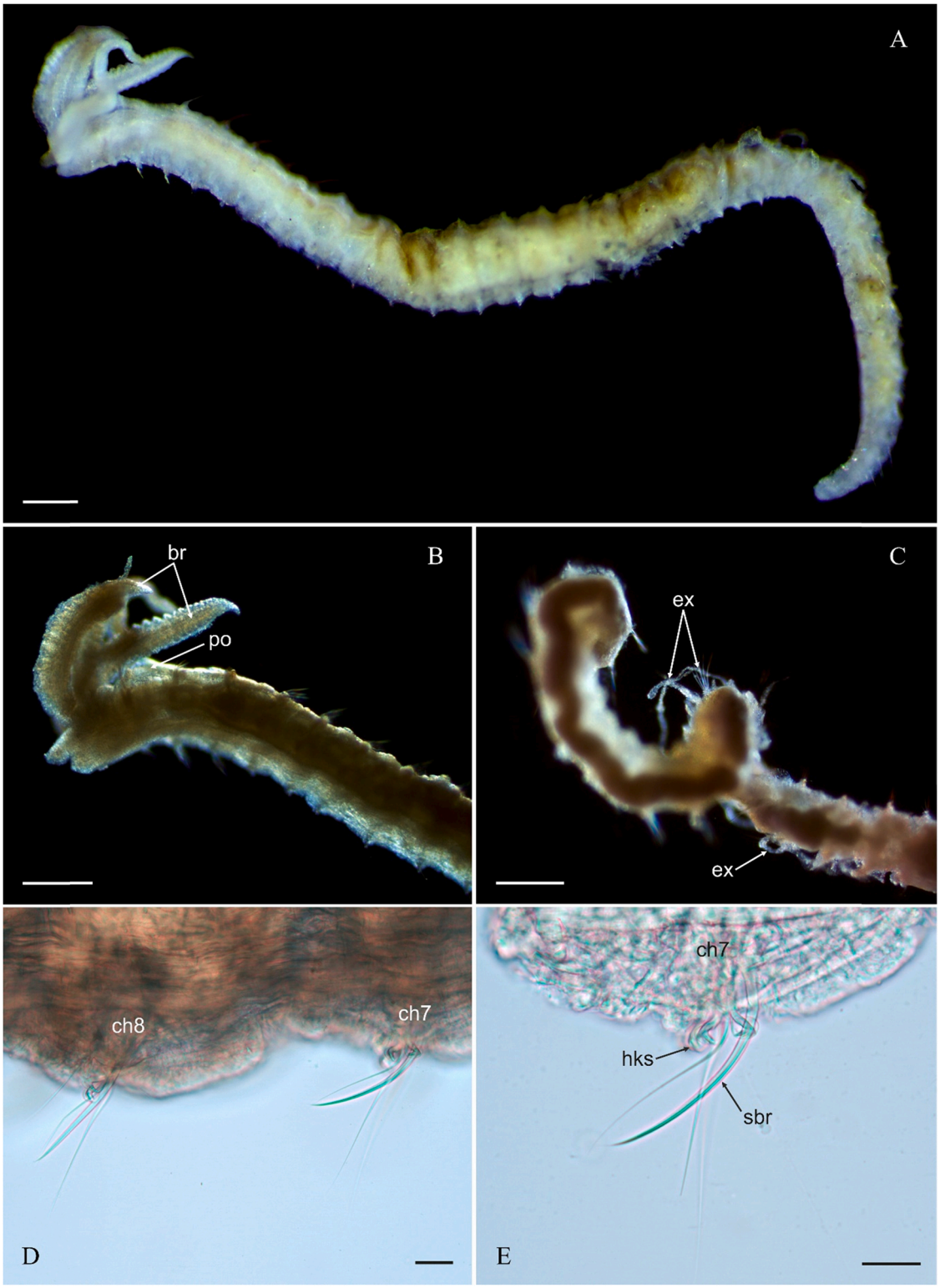

Fig. 2. ***Streblospio gynobranchiata*** **(female) from the Romanian Black Sea** A - Complete specimen, left lateral view; B – anterior end left lateral view; C - middle chaetigers with extensions of lateral body wall; D, E – chaetigers 7 – 8, lateral view (br – branchia; ch7-ch8 – chaetigers 7 – 8; hks – multidentate hooks in neuropodia; ex - extension of lateral body wall; po - dorsal transverse pouch on chaetiger 2; sbr – sabre chaetae). Scale bars: A, B, C = 200 μm, D, E = 20 μm.

The Northern unit (especially the Sulina - Sf. Gheorghe section) is more strongly influenced by the southward longshore current (Duţu et al., 2018), being one of geomorphologically dynamic of the Romanian coastline. However, the natural flow is disrupted by the Sulina jetties, giving rise to a sheltered environment just behind.

The Musura Spit and Bay, at the Stary Stambulsky distributary mouth, and the Sakhalin Island, at the Sf. Gheorghe mouth, have a rapid development in front of the large river discharge mouth (Panin and Overmars, 2012). Musura Bay became shallower due to the constant freshwater and sediment supply from the southern branches of the Kilia Delta (i.e., Stary Stambulsky, Musura and Lebedinoe, as well as other smaller distributaries), which will lead to its closure in near future by a sandy spit that grows over 200 m/year. Southward of the Sf. Gheorghe mouth, a spit island called Sakhalin emerged in 1897, after an exceptional flood, and continued to stretch towards South-West to reach today over 17 km in length (Panin and Jipa, 1998; Panin and Overmars, 2012), while displaying backstepping migration westward through overwashing (30–70 m/year) (Panin and Overmars, 2012). The Sf. Gheorghe meander loops cut-offs (1981–1992 period) triggered an increase of about 6 – 8% in the distributary water and sediment discharge, leading to a more stable coastal sedimentary budget and to a fast development of the Island (Tiron Dutu et al., 2019). Musura and Sakhalin Bays will suffer a gradual transition from marine to a semi-enclosed brackish lagoons and later into freshwater lakes (Stănică et al., 2007).

The water temperature was 27.1 °C in Musura Bay and 26.8 °C in Sakhalin Lagoon, while the surface water salinity was 0.89‰ and 9.1‰, respectively and the dissolved oxygen concentration of surface water was of 10.2 mg/L and 5.76 mg/L, respectively (Pavel et al., 2019).

Fresh water inputs form an upper layer over the saline Mediterranean waters, giving rise to a distinct and permanent pycnocline between the surface (upper 150–200 m) and the deep waters, that limits the vertical exchange and creates a unique chemical and biological oxic/anoxic environment (Konovalov et al., 2005). As much as 87% of the Black Sea is entirely anoxic and contains high levels of hydrogen sulphide (Zaitsev and Mamaev, 1997). Romanian continental shelf waters are characterized by strong spatial and seasonal changes in temperature (0°–27 °C) and salinity (4 – 17‰) under Danube River and wind influences (Berlinsky et al., 2006). The average salinity in the Black Sea is about 17–18 psu in surface waters and 22 in deep waters.

The Southern unit (from Cape Midia up to the Romanian-Bulgarian border) suffers more wind driven changes in physical parameters along the water column, both in cold an warm season. Thus, salinity (a limiting factor for NIS dispersion), is related with NE and SW wind direction. Due to the predominant NE wind, the Danube discharge is kept closer to the coast. The temperature and salinity gradients determine distortions of the surface currents and winds (Dinu et al., 2011).

The Southern unit (from Cape Midia up to the Romanian-Bulgarian border) is more stable and suffers more anthropogenic pressures. This sector is characterised mainly by the presence of an almost continuous loess cliff (12–20 m high), which is more stable as compared with the northern, deltaic coast (Panin and Jipa, 1998). The shores, south of Constanta, do not receive Danube sandy sediments, so that shore deposits are typically of organic origin, mainly derived from mollusc shells the mechanical workout.

Five biological benthic zones are distinguished in the Black Sea: infralittoral, circalittoral, offshore circalittoral, bathyal and abyssal. The substrate type and the wave kinetic energy (2.9 N/m^2) is used to delineate the soft bottom infralittoral from the circalittoral, the constant sea-bottom temperature (9.7 °C on the about 50 m depth) - between circalittoral and offshore circalittoral and density (Sigma-t) for the classification into oxygen regimes and delineation between oxic (>15.9 kg m^{-3}) and suboxic offshore circalittoral (<16.4 kg m^{-3}), while the break of slope - to separate bathyal of abysal (Populus et al., 2017; Vasquez et al., 2020). The study area in the marine environment lies within the infralittoral and circalittoral bionomic zone.

2.2. Data collection and analysis

Field collections were made across the entire Romanian coast of the Black Sea from 2011 to 2020 on 41 marine and 9 brackish (Musura Bay, Sakhalin area) stations (Supplementary Material and Fig. 1). Field activities were performed within National CORE Program of Monitoring of Marine Environment. Five transects were established (Sulina, Sf. Gheorghe, Portita, Constanta and Mangalia). Beside this, 35 of samples were collected in Natura 2000 sites (ROSCI066 – Danube Delta marine part, ROSCI0273 - Marine area of Cape Tuzla, ROSCI0281 – Cape Aurora and ROSCI0094 - Submarine sulfur springs from Mangalia). The time period cover all season in the years 2011–2020.

Additional material from the Danube Delta marine lagoon system (ROSCI0065) was collected through the CORE Program in August 2018 (13 N/2018 – PN 18 16 01 02), while the polychaetes from nine stations at Musura Bay and Sakhalin Lagoon were provided by Dr. Pavel Ana-Bianca.

Single replicates of macrobenthic samples were collected using van Veen grabs with grasping areas of 0.135 m^2 and 0.125 m^2, respectively, on board of the R/V "Mare Nigrum". Samples were washed with sea water using a 0.5 mm mesh sieve (Begun et al., 2018; Wijsman et al., 1999; Bacescu et al., 1971; Todorova and Konsulova, 2005) and the retained organisms were fixed in 4% neutralized formalin in seawater solution. Density and biomass (as wet wight) were referred to one square metre (indv.m^{-2}, g.m^{-2}).

The polychaetes were subsequently preserved in 70% ethanol for morphological and biometric analysis, which were carried out under a Zeiss SteREO Discovery.V8 stereomicroscope, AxioLab.A1 transmitted light microscope and an AxioVert.A1 inverted microscope with DIC technology. The following diagnostic characters were measured: length of branchial crown, length of trunk, width, number of thoracic chaetigers, number of abdominal chaetigers and number of branchial filaments (radioles). Unstained specimens were used for full body and diagnostic character micrographs, taken with a Zeiss Axiocam 305 color digital camera. Measurements were taken with the Zeiss Image software (ZEN 2.5 – blue edition) with licensed Measurement module. Plates were prepared with the CorelDRAW 2017 software. Best preserved and complete specimens (representative of the variability among individuals) were measured and thoroughly analysed. All the material was deposited at the National Research-Development Institute for Marine Geology and Geoecology – GeoEcoMar.

The spatial data distribution was projected using Ocean Data View, version 5.4.0 software (Schlitzer, 2018). Univariate analysis was performed using PRIMER 7 and PERMANOVA+ software package program, version 7.0.17. Species nomenclature was checked according to the World Register of Marine Species (WoRMS Editorial Board, 2021).

Bottom water layer physico-chemical parameters were measured using YSI EXO2 multiparameter probe device. Folk 5/7 was used for sediment classification of the marine habitat delineation according to the EMODnet Seabed Habitats substrate and national classification schemes (Populus et al., 2017; Vasquez et al., 2020).

3. Results

3.1. Environmental parameters

3.1.1. Marine environment (Black Sea)

The marine area comprises all type of sediment classes varying with the depth gradient and extent of the plume area of Danube, although it was dominated by mud (71%) (Appendix A).

The temperature in the near-bottom seawater layer varied from 6° to 19.6 °C and salinity – from 14.8‰ to 18.3‰, according to the season, with the lowest value being measured in March 2017 and the maximums in autumn of 2014 and 2019 (Appendix A).

3.1.2. Estuarine environment

The bottom substrata in the Musura Bay and Sakhalin Lagoon were characterised by muddy (silty clay) sediments. Musura Bay is a shallow semi-enclosed bay (about 1–2 m) with low salinity (1–5‰). The water quality parameter (temperature, salinity and dissolved oxygen) in this study were within the limits set by the Order 161/2006 (Appendix A).

3.2. Benthic habitats and communities

Eight Broad Scale Habitats have been identified in the studied area:

1. Infralittoral fine sand dominated by *Lentidium mediterraneum* and/or *Cerastoderma glaucum* (MB54).
2. Infralittoral fine and medium sand dominated by *Chamelea gallina* and/or *Macomangulus tenuis* (MB54).
3. Lower infralittoral sand and muddy sand with *Upogebia pusilla*, *Micronephthys longicornis*, *Prionospio maciolekae*, *Nephtys hombergii* and *Chamelea gallina* (MB543).
4. Circalittoral mud and sandy mud with *Upogebia pusilla* (MC64).
5. Danube plume area (Mud with *Melinna palmata*, *Mya arenaria*, *Anadara kagoshimensis*, *Alitta succinea*, *Nephtys hombergii*) (MC641).
6. Circalittoral mud dominated by *Melinna palmata* (MC64).
7. Circalittoral mud with *Spisula subtruncata*, *Abra nitida*, *Pitar rudis*, *Acanthocardia paucicostata*, *Nephthys hombergii* and *Mytilus galloprovincialis* beds (MC644).
8. Circalittoral mixed sediments with varied infauna and musselbeds of *Mytilus galloprovincialis* (MC241).

The marine benthos includes six major invertebrate taxonomic groups, with the Polychaeta being dominant in number of species and abundance, followed by Oligochaeta and Mollusca, a low representation of Nemertea, Phoronida and Crustacea.

The estuarine benthos is dominated by tube-dwelling corophiids (Amphipoda), followed by Polychaeta, Oligochaeta and larvae of Chironomidae, most of them typical from fresh- and brackish-water environments.

4811 individuals of *Streblospio gynobranchiata* and *Laonome xeprovala* were found at the Romanian Black Sea Coast, 4691 from the marine area and 120 from the estuarine area.

3.3. Systematics

3.3.1. *Streblospio gynobranchiata* Rice and Levin (1998) (Fig. 2)

3.3.1.1. Material examined. **Romania, NW Black Sea**, 8 April 2011, MA05, 86 ind; 23 August 2012, TZ17, 2 ind; 27 August 2013, W09, 75 ind; 16 October 2014, 20 C, 1 ind; 22 September 2014, 2PII/S3, 1 ind; 2PII/S4, 2 ind; 22 February 2015, 3PII/S1, 1 ind; 16 April 2015, PO02, 4 ind; 10 August 2015, PO02, 1 ind; 11 August 2015, SU01, 11 ind; 18 November 2015, TZ10, 2 ind; TZ14, 1 ind; 19 November 2015, MG13, 2 ind; 19 May 2016, NPMS UA07, 22 ind; 3 July 2016, 4, 2 ind; 27 March 2017, CA03, 1 ind; 17 August 2018, PO01, 1984 ind; PO02, 93 ind; SG01, 130 ind; 18 August 2018, SU01, 10 ind; 24 August 2018, MA06, 1 ind; 25 August 2018, EF02, 1 ind; 26 August 2018, CT01, 3 ind; CT02, 7 ind; 20 August 2019, S09, 1 ind; 2 September 2019, SU01, 1 ind; 16 October 2019, PI-S1, 3 ind; PI-S2, 6 ind; 17 October 2019, PII-S1, 3 ind; PII-S2, 324 ind; PIII-S1, 37 ind; PIII-S2, 57 ind; 18 October 2019, PIV-S1, 11 ind; PIV-S2, 28 ind; PV-S1, 9 ind; PV-S2, 79 ind; 18 September 2020, PO01, 1602 ind; 19 September 2020, SG01, 57 ind; SG03, 2 ind; 20 September 2020, SU01, 27 ind; SU03, 1 ind.

3.3.1.2. Additional material (coll. A.B Pavel). Musura Bay, August 2018, DD18–157, 10 ind; Sakhalin lagoon, August 2018, DD18–171, 6 ind; DD18–173, 10 ind; DD18–174, 12 ind; DD18–175, 20 ind; DD18–176, 12 ind; DD18–176 N, 25 ind.

3.3.1.3. Short morphological description. Up to 6 mm long (4.8 mm on average) and 0.3 mm wide (0.27 mm on average) for 42–45 chaetigers (Table 2). Prostomium elongate, anteriorly round.

Peristomium forming a prominent hood enveloping prostomium laterally and ventrally. Two pairs of black small eyes in trapezoidal arrangement. One pair of palps on midlateral side of prostomium, as long as 6–7 chaetigers. Median papilla on dorsal side of chaetiger 1. Chaetiger 1 with short capillaries in both rami and small postchaetal lamellae in neuropodia; notopodial lamellae absent. Characteristic dorsal transverse pouch open anteriorly on chaetiger 2. Sabre chaetae and multidentate hooded hooks in neuropodia from chaetiger 7. Single pair of branchiae on chaetiger 1, posterior to palps. Branchiae apinnate, with undulating lateral edges, almost equal in length to palps. Finger-like extensions of lateral body wall in mature females from chaetigers 17–18. Pygidium simple, small, round, without anal cirri.

3.3.1.4. Ecology. The average density ranged from 636.4 ind.m^{-2} (2011) to 2702.4 ind.m^{-2} (2020). From 2014–2017, the density ranged between 7.4 ind.m^{-2} and 162.8 ind.m^{-2}, but the more extensive study from the last three years clearly indicates that this opportunistic species was occupying new biotopes along the Romanian coastline.

The highest densities (4945.2 ± 7599.4 ind.m^{-2}) were recorded at 11–15 m depth in the infralittoral sandy mud, and the lowest (10.2 ± 3.8 ind.m^{-2}) at 31–36 m in circalittoral mixed sediments and circalittoral mud with *Melinna* and *Spisula* habitats (Figs. 3 and 4).

In the Northern unit *S. gynobranchiata* is more frequent and abundant than in the Southern unit (Figs. 3 and 5), with the highest density (3765.2 ± 6788.5 ind.m^{-2}) being found at Portita, followed by Sakhalin Lagoon (3440.7 ± 2543.2 ind.m^{-2}).

3.3.2. Laonome xeprovala Bick & Bastrop, in Bick et al. (2018) (Fig. 6)

3.3.2.1. Material examined. **Romania,** Musura Bay, August 2018, DD18-156, 2 ind; Sakhalin Lagoon, August 2018, DD18-176, 34 ind.

3.3.2.2. Short morphological description. Body without branchial crown, 4–27 mm long and the width ranged from 0.4 to 1.6 mm wide (Table 3) with eight thoracic (in all adults) and 22–46 abdominal (depending on length) segments. Branchial crown 1.1–3.7 mm long (i.e., 18% of total body length on average in specimens > 10 mm and 27% when < 10 mm). 4–10 pairs of pinnulate branchial radioles crossed by 5–7 transverse light-brown bands; pinnules arranged in two alternating rows. Gill membrane present. Transverse lamellar process (ridge) present at the junction between peristomium and chaetiger 1.

First notopodium with elongate narrowly-hooded chaetae, then chaetal fascicles with 3–6 hairy-bordered capillaries and 5–8 paleate chaetae (arranged in a single row) chaetae. Abdominal chaetigers with about 5–8 broadly hooded neurochaetae arranged in a single row and 10–22 notopodial avicular-type uncini with several rows of teeth over main fang. Thoracic and abdominal uncini lacking handles. Pygidial eyes absent. Tube wall formed by mucus agglutinating fine sediment particles.

3.3.2.3. Remarks. The morphologic characters cleary allow to identify the specimens as belonging to *Laonome* Malmgren, 1866, particularly the transverse lamellar process (ridge) at the junction between peristomium and chaetiger 1 and the absence of handles in thoracic and abdominal uncini (Boltacheva, 2017; Bick et al., 2018).

3.3.2.4. Ecology. Laonome xeprovala was been found from 0.5 to 1.5 m depth in the brackish coastal lagoons at Musura Bay (sandy mud) and Sakhalin Lagoon (sand).

We found both adults and juveniles, although the former were dominant. Its density ranged from 7.4 ind.m^{-2} (Musura Bay) to 100.8 ind.m^{-2} (Sakhalin Lagoon).

Table 2
Length and width of the *Streblospio gynobranchiata* population from the Romanian Black Sea and adjacent waters.

No. crt.	Length [mm]	Width [mm]	No. crt.	Length [mm]	Width [mm]
1	3.634	0.22	13	4.527	0.27
2	4.084	0.22	14	4.597	0.261
3	4.1	0.29	15	5.262	0.28
4	4.105	0.23	16	5.285	0.28
5	4.167	0.33	17	5.3022	0.28
6	4.21	0.231	18	5.432	0.26
7	4.287	0.28	19	5.467	0.272
8	4.294	0.28	20	5.782	0.4
9	4.294	0.253	21	5.891	0.279
10	4.37	0.265	22	5.891	0.307
11	4.43	0.241	23	5.92	0.364
12	4.451	0.3	24	5.992	0.26

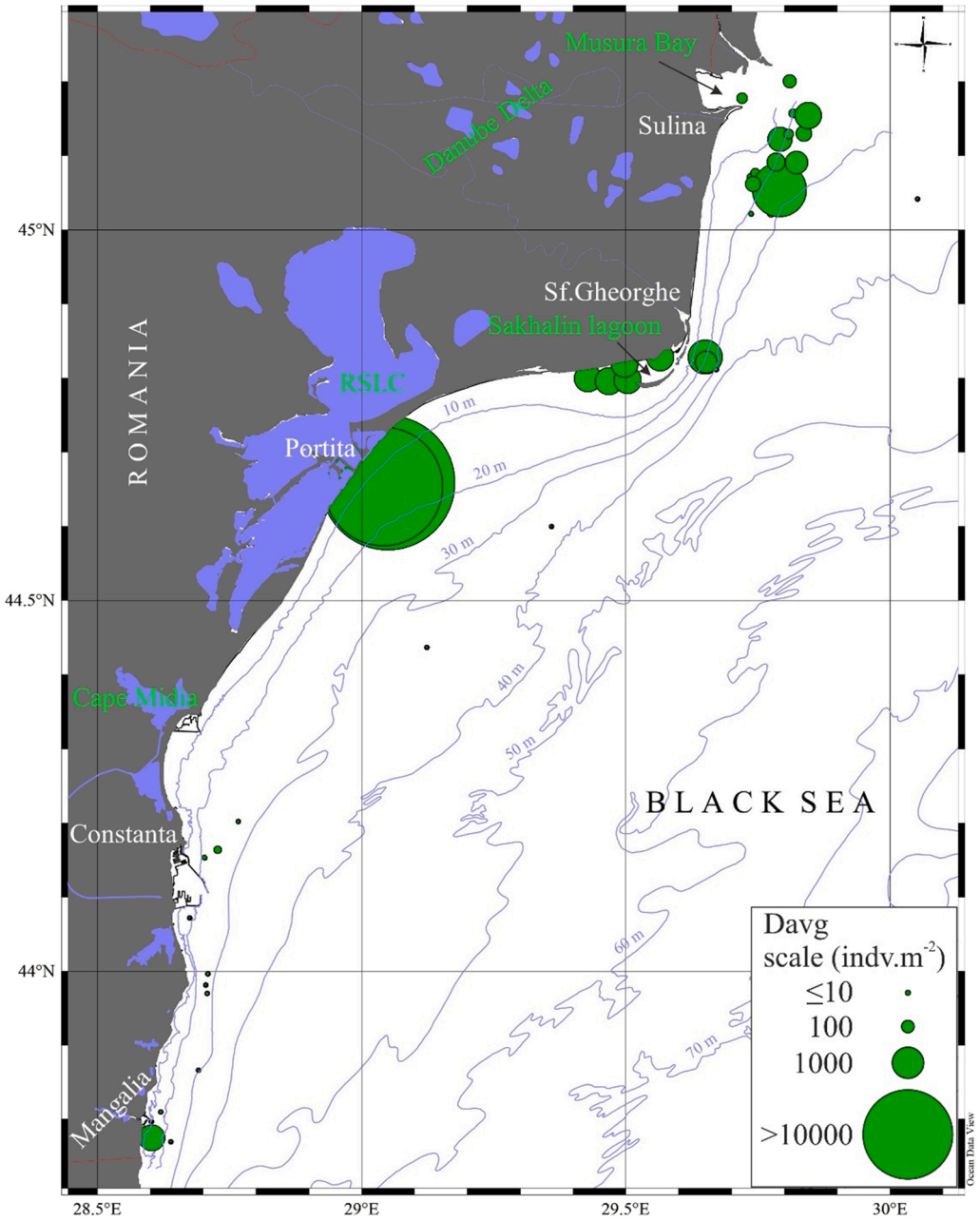

Fig. 3. Average abundance of *S. gynobranchiata* along Black Sea Romanian coast.

4. Discussion

4.1. Streblospio gynobranchiata

Spionidae is one of the most abundant and diverse families of polychaetes, occurring in almost all marine habitats from shallow waters to the deep-sea a calcareous substrates and rock crevices to a wide diversity of sedimentary habitats throughout all world oceans (Blake, 1996). Many spionids are euryhaline opportunists inhabiting a large variety of stressed environments, which make them useful indicators (Pearson and Rosenberg, 1978; Ritter and Montagna, 1999; Surugiu, 2005). Among them, the species of *Streblospio* occurs from West (Pearse et al., 1987) to East Atlantic Ocean, as well as in the Mediterranean Sea (Sarda and Martin, 1993). Currently, in addition to *S. gynobranchiata*, seven species are considered as valid: *S. benedicti* Webster, 1879, *S. shrubsolii* Buchanan, 1890, *S. japonica* Imajima, 1990, *S. padventralis* Delgado-Blas, Díaz-Díaz & Viéitez, 2018, *S. eunateae* Martínez and Adarraga, 2019, and *S. eridani*

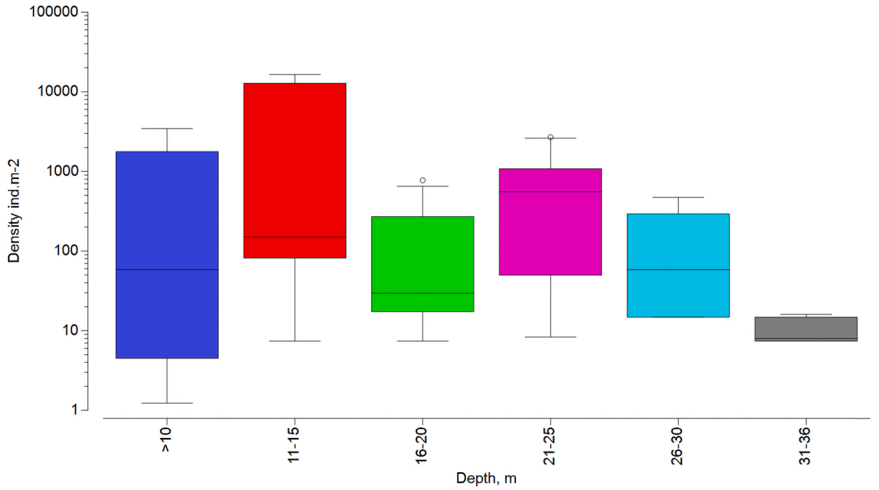

Fig. 4. Bathymetric distribution of density of *S. gynobranchiata* at the Black Sea Romanian coast.

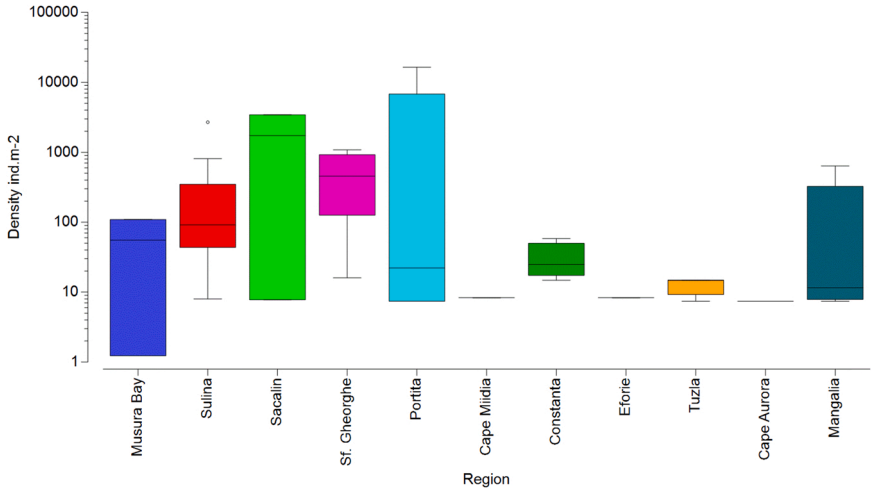

Fig. 5. Geographical variability of density of *S. gynobranchiata* along the Black Sea Romanian coast.

Munari, Wolf, Infantini, Moro, Sfriso & Mistri, 2020, but only two (*S. shrubsolii* and *S. gynobranchiata*) occur in the Black Sea (Marinov, 1957; Boltacheva, 2008; Radashevsky and Selifonova, 2013).

Streblospio gynobranchiata was first observed in soft sediment from the Mangalia, in April 2011 at 17 m depth, with bottom water salinity of 18‰, and temperature of 7.90 C and muddy sediments. The average density and biomass were 636.4 ind.m^{-2} and 0.191 g. wwt.m^{-2} (Teaca et al., 2015). Since then, we have been commonly observing the species in benthic samples, but it was not been mentioned by other Romanian institutes. However, this species forms dense agglomerations in Romanian Black Sea soft bottoms, were it has been recently introduced (Teaca et al., 2015), so that our data are among the first known on the spatial distribution in Romanian coasts. The specimens collected for this study agree with the original description of *S. gynobranchiata* by Rice and Levin (1998) based on specimens from Tampa Bay (Florida, USA), particularly in having a single pair of branchiae and lateral extensions in the body wall. Later, it was reported from the Aegean Sea, Turkey (Çinar et al., 2005a, 2005b, 2012; Dağli et al., 2011), Marmara Sea, Turkey (Çinar et al., 2009), Novorossiysk port of the Black Sea (Murina et al., 2008), Crimea in the northern Black Sea (Boltacheva, 2008; Boltacheva et al., 2021), Sukhoi Liman NW Black Sea (Radashevsky and Selifonova, 2013), Mangalia port of the Black Sea (Teaca et al., 2015), the southern part of the Caspian Sea (Taheri et al., 2008; Taheri and Foshtomi, 2011), and the Sukhum Bay of the Black Sea (Selifonova and Bartsits, 2018) (Fig. 7). Our data confirm the presence of the species in Romanian waters, with the north sandy and muddy coast (which highly influenced by freshwater inputs and sediment inflow) showing the maximum densities (ca. 17,000 ind.m^{-2}). Conversely, in the southern coast (which is characterized by a mosaic of sediments) densities were much lower ($<$ 500 ind.m^{-2}) and the species was only present in the stations near the ports of Constanta and Mangalia, with an average of 68.1 $\pm$ 179.1 ind.m^{-2}.

Streblospio gynobranchiata is a temperate tube-dwelling, semi-sesil infaunal spionid, often very abundant in muddy sands (Rice and Levin, 1998), usually inhabiting the upper sediment layer. With the help of its ciliated tentacles and like other species of *Streblospio* it

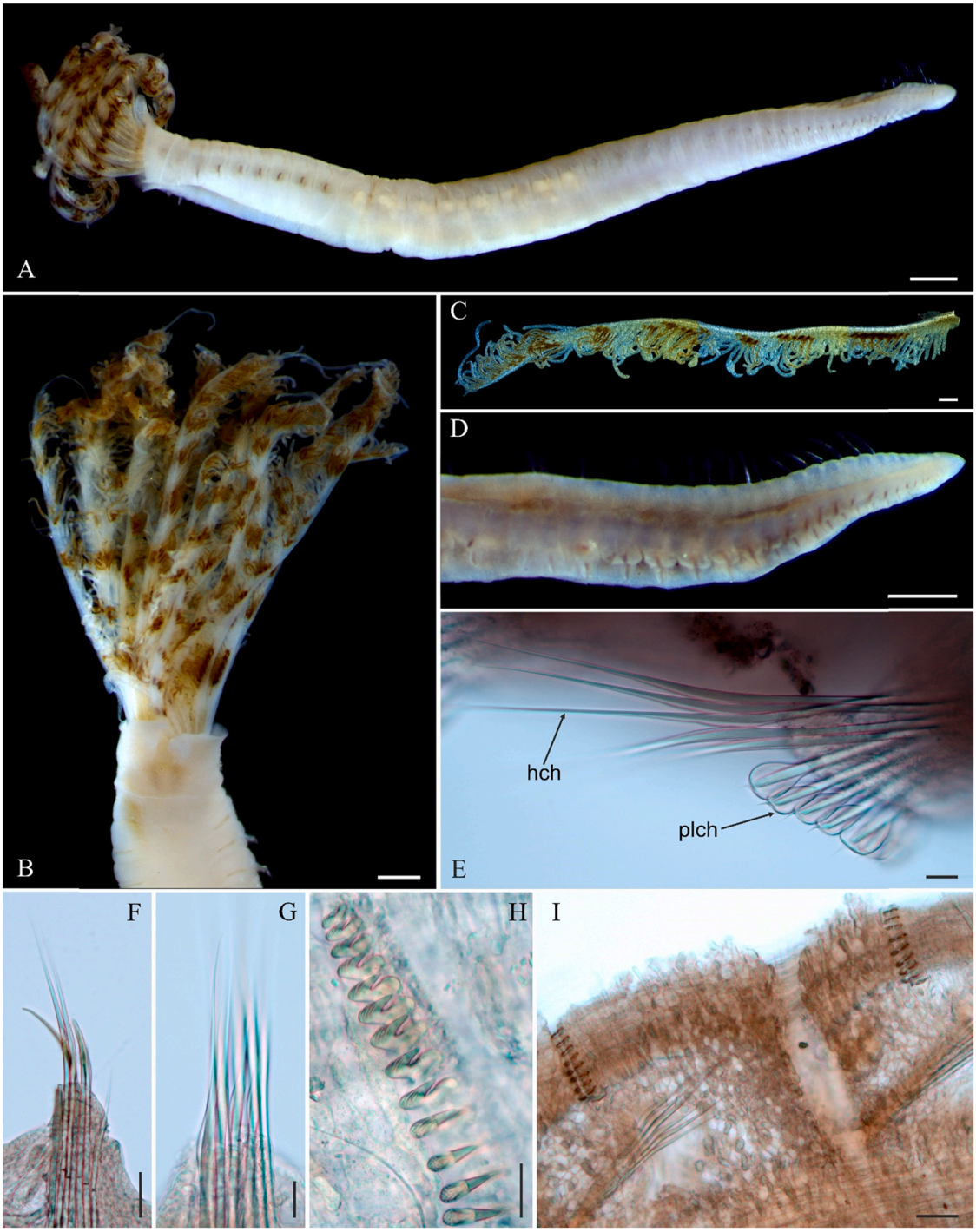

Fig. 6. ***Laonome xeprovala*** **from Sakhalin Lagoon, Black Sea**. A – Complete specimen; B – anterior end with enlarged branchial crown; C – branchial radiole with characteristic brown transverse bands; D - posterior end - pygidium and anal depression; E - fourth notopodium with five superior capillary unilimbate chaetae and seven inferior paleate chaetae; F, G – first notopodium (from the same specimen) with one acicular spine and 6 narrowly hooded chaetae and without inferior paleate chaetae; H – a row of 13 abdominal uncini; I - abdominal chaetigers with six broadly hooded neurochaetae and ten notopodial avicular-type uncini (*hch* – narrowly hooded chaetae; *plch* - paleate chaetae). Scale bars: A, B, D = 500 μm, C = 200 μm, E, G, H = 20 μm, F, I = 50 μm.

Table 3
Biometric characteristics of *Laonome xeprovala* recorded in the Black Sea Romanian coast and adjacent waters.

No. crt.	Length [mm]	Width [mm]	Branchial crown [mm]	No. of thoracic segments	No. of abdominal segments	No. of branchial radioles
1	16.426	0.728	2.445	8	39	16
2	16.86	0.632	2.907	8	38	16
3	13.938	0.62	2.551	8	38	14
4	17.963	0.676	3.753	8	41	18
5	19.383	0.87	3.762	8	41	16
6	17.076	0.834	3.297	8	41	18
7	9.275	0.598	2.53	8	34	12
8	13.789	0.916	2.506	8	38	14
9	16.512	1.031	2.697	8	46	16
10	27.782	0.852	3.656	8	45	18
11	–	0.785	2.703	8	–	14
12	19.632	0.886	2.517	8	46	18
13	–	0.787	2.318	8	–	14
14	12.174	0.663	2.886	8	36	14
15	7.863	0.637	2.507	8	30	12
16	8.557	0.838	2.491	8	35	12
17	8.977	0.819	2.458	8	35	14
18	10.238	0.708	1.992	8	34	14
19	–	0.77	2.571	8	–	16
20	7.6	0.664	1.921	8	33	12
21	12.668	0.645	2.088	8	35	14
22	9.261	0.703	2.591	8	36	14
23	12.123	0.826	2.512	8	34	14
24	6.8	0.637	1.512	8	31	12
25	8.686	0.759	2.214	8	36	12
26	4.096	0.437	1.142	8	22	8
27	–	1.623	4.945	8	–	20
28	–	0.722	2.482	8	–	16
29	–	0.452	1.999	8	–	8
30	8.766	0.614	1.746	8	32	14
31	4.911	0.58	1.777	8	24	8

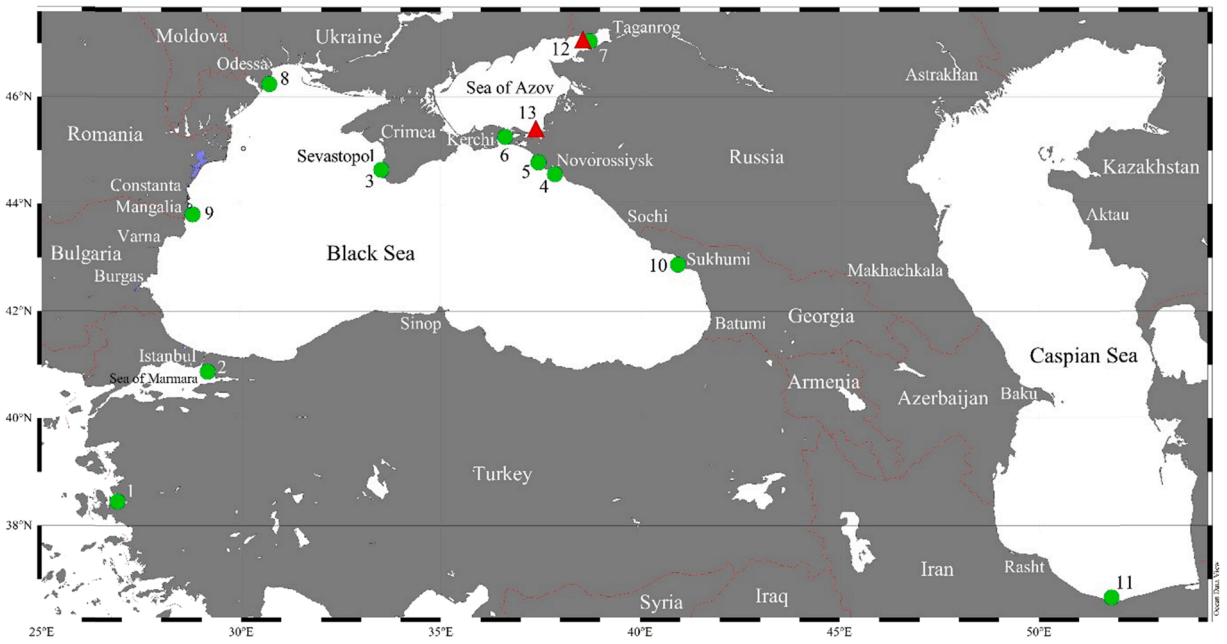

Fig. 7. Known records of *S. gynobranchiata* and *L. xeprovala* in the Eastern Mediterranean, Black and Caspian Seas and the Seas of Azov and Marmara. *Streblospio gynobranchiata* (green dots): 1 - Çinar et al. (2005a), (2005b), Dağli et al. (2011); 2 - Çinar et al. (2009); 3 - Boltacheva (2008), Boltacheva et al. (2015); 4 - Murina et al. (2008), Radashevsky and Selifonova (2013); 5 – Radashevsky and Selifonova (2013); 6 - Boltacheva and Lisitskaya (2019); 7 - Syomin et al. (2017); 8 – Radashevsky and Selifonova (2013); 9 - Teaca et al. (2015); 10 - Selifonova and Bartsits (2018); 11 - Taheri et al. (2008), Taheri and Foshtomi (2011). *Laonome xeprovala* (red triangles): 12 - Syomin et al. (2015), Bick et al. (2018); 13 - Boltacheva et al. (2017). (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

may, filtrate water to feed on seston or, when phytoplankton concentrations are low, alternatively feed on bottom detritus (George and Hartmann-Schröder, 1985; Hartmann-Schröder, 1996; Dauer et al., 1981). This opportunistic species is characterized by its small body and shows a high reproductive capacity, short life span high colonization capacity and high mortality (Dauer et al., 1981). The planktotrophic larvae occur in the plankton from three to the 9–12 chaetiger stages, and settle occurs after 9–10 days (at 17–18 °C) (Rice and Levin, 1998). The reproductive success is highly influenced by food availability, but also by substrate availability, as the absence of suitable bottoms may extend the larval period to one month (Boltacheva et al., 2015).

Streblospio gynobranchiata seems to have been accidently introduced in the Aegean, Caspian and Black Seas with ballast water (Çinar et al., 2005b; Taheri et al., 2008; Boltacheva et al., 2021) and we suggest the same for the Romanian Black Sea. We found less than 20 ind.m^{-2} at 36.5 m of maximum depth in circalittoral mud with *Melinna* and *Spisula* habitat, while the highest densities (ca 17, 000 ind.m^{-2} in Portita at 13.2 m depth) were half those of a recruitment area in Alsancak Harbour (Izmir Bay, Aegean Sea) (Çinar et al., 2005b), and ca. three time less than in the Danube - Black Sea navigable channel (5–6 km upstream from shore) (Vinogradov et al., 2018), which showed a much higher primary production and organically enriched superficial sediments (Vinogradov et al., 2018), revealing a positive correlation between, the density of *S. gynobranchiata* and the percentage of total organic matter (Taheri et al., 2008). This species had a great impact on the benthic habitat and replaced some opportunistic species previously known from the polluted Izmir Bay such as *Capitella* sp. (Çinar et al., 2005b). In fact, in the Romanian Black Sea, the *S. gynobranchiata/C. capitata* association rarely occurs, with *C. capitata* seldom exceeding 24 ind.m^{-2}. Conversely, back in the 90 s, *C. capitata* frequently exceeded 12,800 ind.m^{-2} between 20 and 30 m depth (Gomoiu, 1999). Therefore, the lack of successful recruitment, the observed low densities, and, even its absence (particularly offshore) (Muresan, 2021), leads us to conclude that their distribution in the Romanian continental shelf is basically the pollution (i.e. organic enrichment) limited.

Salinity is another factor affecting population success in marine polychaetes, by limiting their spatial distribution. In River Tsemess estuary (Novorossiysk Harbour), adults occur at 12.9‰ (Radashevsky and Selifonova, 2013) and at Izmir Bay (Eastern Mediterranean) at 35.1–39.8‰ (Çinar et al., 2008), while in our study region, salinity ranged between 14.3‰ and 17.8‰ in the marine sectors and from 2‰ to 10‰ in the estuarine area. The abundance of *S. benedicti* decreases together with salinity decline (Reish, 1979) and *S. gynobranchiata* most likely exhibits a similar response. Nevertheless, more *S. gynobranchiata* was found in high density (3440 ind. m^{-2}) at Sakhalin lagoon, with surface water salinities of 2.5–10‰ in agreement with Vinogradov et al. (2018). Therefore, *S. gynobranchiata* seemed to have salinity tolerance wider than previously assumed, as well as a high capacity to adapt to new conditions such as estuarine habitats. Overall, it took less than 10 years for this species to spread along the entire Romanian Black Sea coast on different types of substrata and habitats, being influenced by different environmental factors and apparently lacking any dispersal limitation.

4.2. *Laonome xeprovala*

The sabelid *L. xeprovala* had not been reported in the Black Sea to date. Thus our findings represent its first report for a Black Sea estuarine environment.

There are ten known species of *Laonome* Malmgren, 1866 and, so far, seven species from seven genera of Sabellidae have been reported in the Black Sea (Surugiu and Capa, 2020). *Laonome xeprovala* was reported in the Baltic Sea (Capa, 2014; Kotta et al., 2015; Kocheshkova, 2017; Bick et al., 2018; Kocheshkova and Ezhova, 2018) and in the adjacent Sea of Azov (Syomin et al., 2015; Boltacheva et al., 2017) (Fig. 7). Introduction pathways of seemed to be through shipping and ballast waters from the North Atlantic and the Baltic Sea via the Volga-Baltic waterway and the Volga-Don channel (Boltacheva et al., 2017) down to Sea of Azov and the Caspian Sea and probably to the adjacent waters of the Danube Delta. However, assuming that *L. xeprovala* shows a similar development to its most closely related species, in which settling of lecithotrophic larvae occurs within 26–33 h after fertilization, their larval stage probably have a low capacity for long distance dispersal. Anyway, the larvae may survive in ballast tanks and, after released, settle on substratum as post-larval (Capa et al., 2014).

Laonome xeprovala is a freshwater and brackish water tube-dwelling suspension feeder, occurring in hard bottoms, sandy silt with detritus, clean sand, soft clay rich in organic material and decomposing microalgae (Bick et al., 2018; Tamulyonis et al., 2020) and mixed se diments (Boltacheva et al., 2017; Bick et al., 2018) from 0.5 to 17 m depth (Boltacheva et al., 2017; Bick et al., 2018; Tamulyonis et al., 2020).

In the Baltic Sea, *L. xeprovala* seemed to be predominantly associated to organically enriched areas with coarse-detritic silts (Kotta et al., 2015; Kocheshkova, 2017; Kocheshkova and Ezhova, 2018; Tamulyonis et al., 2020), which probably facilitates its development and support its adequate nutrition. We have observed this species on sandy mud and sandy sediments, with 100 ind.m^{-2} or less, while they reach more than 1300 ind.m^{-2} in the Baltic Sea (Tamulyonis et al., 2020) and around 400 ind.m^{-2} in Sea of Azov (Boltacheva et al., 2017). This species rapidly dominates new habitats, potentially modifying the sediment morphology and chemistry and disrupting the native benthic communities (Bick et al., 2018). Also, as eurybiont, it has a high potential to expand its distribution range, especially along different ranges of salinity and temperature. In our study area, we have not yet detected any visible effect on benthic habitats. However, the population will turn to be stabilize over time, thus, it would also be possible to quantify the effects on native benthic faunal assemblages. To date, the species just occupies empty ecological niches at the Musura Bay and Sakhalin Lagoon, thus artificially increasing the respective species richness.

5. Conclussions

The finding of *L. xeprovala* and *S. gynobranchiata* contribute to improve our knowledge on the diversity of Black Sea and adjacent

areas, although further efforts must be done in order to obtain a broad picture of their impact in such a threatened ecological system.

Once a new species has turned up in the environment, it brings about ecosystem changes that can be both positive and negative. In the Black Sea, we do have examples of NIS succeeding to extend their distribution at the expenses of native species (e.g., *Dipolydora quadrilobata*), while others that have either an uncertain status due to their rare occurrence (i.e., *Sigambra tentaculata*) or just occur in specialised niches (i.e., *Hydroides dianthus*). Due to their biological features and wide ecological tolerance range, *L. xeprovala* and *S. gynobranchiata* may also be successfully established in the Black Sea. Nevertheless, the accelerated ecological changes (warming, hypoxia, NIS incoming, anthropogenic pressure) may act as spreading limiting factors.

Funding

This study was supported by the Ministry of Research, Innovation and Digitization, Romania in the framework of the CORE Programme projects: PN 19200302, PN 19200401, PN 19200101 and PN 19200102.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

The authors would like to thank all colleagues who helped with samples sorting and made suggestions for manuscript improving, as well as to the anonymous reviewers.

Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.gecco.2021.e01920.

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