Records of Polydora cornuta and Streblospio gynobranchiata (Annelida, Spionidae) from the Black Sea

RADASHEVSKY V.I. A.V. Zhirmunsky Institute of Marine Biology, Far Eastern Branch of the Russian Academy of Sciences, Vladivostok 690059, Russia Far Eastern Federal University, Vladivostok 690091, Russia

SELIFONOVA Z.P. Admiral Ushakov Maritime State University, Novorossiysk 353918, Russia

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Records of *Polydora cornuta* and *Streblospio gynobranchiata* (Annelida, Spionidae) from the Black Sea

V. I. RADASHEVSKY1,2 and Z. P. SELIFONOVA3

1 A.V. Zhirmunsky Institute of Marine Biology, Far Eastern Branch of the Russian Academy of Sciences, Vladivostok 690059, Russia
2 Far Eastern Federal University, Vladivostok 690091, Russia
3 Admiral Ushakov Maritime State University, Novorossiysk 353918, Russia

Corresponding author: radashevsky@gmail.com

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Abstract

Two spionid polychaetes, *Polydora cornuta* and *Streblospio gynobranchiata*, were identified in benthic samples collected in the northern Black Sea and adjacent waters. These species have earlier been classified as the worst invaders in soft bottom communities in the Mediterranean Sea. *Polydora cornuta* had been previously misidentified and widely reported from the Black Sea and the Sea of Azov as *P. ciliata, P. ciliata limicola* and *P. limicola*. *Streblospio gynobranchiata* is a new invader currently extending its distribution into the Mediterranean, Black and Caspian Seas. Morphology, diagnostic characters and biology of the species are discussed and the history of their records in the Mediterranean, Black and Caspian Seas and the Sea of Azov is reviewed.

Keywords: *Polydora cornuta, Streblospio gynobranchiata*, biological invasions, Russia, Ukraine.

Introduction

In the last few decades marine biological invasions caused by human activities and global climate changes have become grave matters of concern due to their ecological and economic ramifications, causing intensified research efforts in this field (Occhipinti-Ambrogi & Savini, 2003; Carlton, 2009; Katsanevakis et al., 2013). A total of 986 alien or non-indigenous species have been recognized in the Mediterranean basin, of which 560 (57%) species have been reported as established (Zenetos et al., 2012). In the Black Sea, about 160 alien species have been identified since the first scientifically documented invasions of the barnacles *Amphibalanus improvisus* (Darwin, 1854) and *A. eburneus* (Gould, 1841) in Sevastopol Bay in 1844 and 1892, respectively (Gomoiu & Skolka, 1996; Zaitsev & Öztürk, 2001; Leppäkoski et al., 2009). A six fold increase in the number of recognized alien species in the Black Sea in the last decade has resulted from not only an increased number of studies but also from the rapid growth of human activities in the region (Alexandrov, 2004).

Lists of marine alien species always include polychaete annelids, among which spionid polychaetes (Annelida: Spionidae) are common invaders (Çinar et al., 2005a, 2011; Sahin & Çinar, 2012). For example, intensive studies of polychaetes along the Turkish coast of the Aegean Sea revealed a total of 35 spionid species of which nine were recognized as alien (Dağlı et al., 2007, 2011). The massive development of *Polydora cornuta* Bosc, 1802 in Odessa Harbour in the early 1960s (Losovskaya & Nesterova, 1964, as *P. ciliata limicola* Annenkova, 1934) was probably one of the most conspicuous marine invasions in the Black Sea. Since then, *P. cornuta* has been reported from harbours in various countries surrounding the Black Sea and is now considered to be one of the worst invasive species in soft bottom communities in the Mediterranean Sea (Zenetos et al., 2005). Another spionid on the list of worst invaders, *Streblospio gynobranchiata* Rice & Levin, 1998, was recently introduced and is currently spreading in the Mediterranean, Black and Caspian Seas.

Two important Russian international seaports, Novorossiysk and Tuapse, are located in the northeast Black Sea (Fig. 1). Their annual traffic capacity is about 5000 and 1000 ships per year, respectively, and their carrying capacity is about 100 and 20 million tons of cargo, respectively. The whole region is therefore subjected to high levels of pollution, ballast water discharge (more than 50 million tons per year in Novorossiysk Bay) and species movement via ship fouling, all factors that favour the introduction and establishment of non-indigenous species. As a consequence, 23 alien and 44 cryptogenic species have been reported from the port areas (Selifon-
Sevastopol and Odessa are among the largest seaports in Ukraine in the north and north-western parts of the Black Sea, respectively. Recent sampling in Novorossiysk Bay and the port of Tuapse, and samples previously collected in Sevastopol, Odessa and adjacent areas, provided good material for taxonomic revisions and better insight into the regional distribution of invasive species of Spionidae.

The purpose of this study is to report new records of spionid polychaetes in the northern Black Sea and adjacent waters, re-examine old material and present an overview of the expansion of invasive sponids in the Mediterranean, Black and Caspian seas and the Sea of Azov.

Materials and Methods

Field collections were made in Snake Lake Liman (Big Utrish) (Zmeinoe Ozero Liman, Bolshoi Utrish), Novorossiysk Harbour (Novorossiysk Bay), Tuapse Harbour and Sochi, all in the north-eastern part of the Black Sea, Russia, by Zhanna P. Selifonova in September 2010, August 2011, November 2011, and July 2012. Sediment samples were collected using a Petersen grab (0.04 m$^2$) and screened in the field with a 500 µm mesh sieve. The residue was fixed in 10% formaldehyde solution, rinsed in fresh water, and then transferred to 70% ethanol. Polychaetes were removed from the residue under a stereomicroscope at the Admiral Ushakov Maritime State University, Novorossiysk. Additional material from the Sea of Azov and the Black Sea (Sevastopol Bay, Odessa and adjacent regions) was provided by Marta I. Kiseleva, Ludmila V. Vorobyova, Ivan V. Sinegub, Elena V. Lisitskaya and Natalia A. Boltacheva. Photographs were taken using a Zeiss Axio Imager 7.2 microscope equipped with a digital camera. Specimens were stained with a solution of methylene green in alcohol prior to photography.

Supplementary material on *P. cornuta* from the Aegean Sea, Greece, was provided by Nomiki Simboura. All the material was deposited at the Museum of the Institute of Marine Biology (MIMB), Vladivostok, Russia, and the Natural History Museum of Los Angeles County (LACM-AHF), Los Angeles, U.S.A. Information about the samples is given below along with descriptions of the specimens. The number of specimens in each sample is
given in parentheses after the museum abbreviation and registration number.

**Taxonomic Account**

*Polydora cornuta* Grube, 1850

*Polydora cornuta* Bosc, 1802


**Material**

SEA OF AZOV, R/V Gidrolog, coll. V.P. Zakutsky: sta. 40, 46°12′N, 37°18′E, 25 Jun 1983, MIMB 27148 (3); sta. 54, 46°12′N, 37°48′E, 26 Jun 1983, MIMB 27149 (11); sta. 55, 45°43′N, 36°43′E, 26 Jun 1983, MIMB 27150 (1); sta. 69, 45°33′N, 35°48′E, 29 Jun 1983, MIMB 27151 (3); sta. 74, 45°43′N, 35°12′E, 29 Jun 1983, MIMB 27152 (6).


UKRAINE, northern Black Sea, Crimea: Karkinya Bay, Swan Islands, 45°51.4′N, 33°29.8′E, intertidal, fouling on a concrete pipe, coll. V.A. Grintso, 12 Aug 2005, MIMB 14114 (6); 0.3 m, muddy sand, coll. E.V, Lisitskaya, 14 Aug 2007, MIMB 27202 (3).

Sevastopol Bay: Martyn Bight, 44°37′N, 33°30.5′E, coll. V.I. Radashevsyky, 2 Dec 2005, MIMB 14115 (1); 44°37.09′N, 33°31.41′E, reared in laboratory from larvae collected from plankton in Sebastopol Bay, M.I. Kiseleva, 10 Nov 1988, MIMB 27153 (20). Reared in laboratory from larvae collected from plankton in Karantinnaya Bay, 44°37.02′N, 33°30.25′E, 10 Aug 2006, E.V. Lisitskaya, MIMB 17927 (1). Reared in laboratory from larvae collected from plankton in Balaklava Bay, 44°30.12′N, 33°35.86′E, on 2 May 2006, fixed on 21 Aug 2006, E.V. Lisitskaya, MIMB 17926 (10); collected from plankton on 21 Aug 2006, fixed on 13 Oct 2006, E.V. Lisitskaya, MIMB 17925 (7).

UKRAINE, north-western Black Sea, Odessa area: off Odessa, 46°30′N, 30°50′E, 7–26 m, muddy sand, coll. I.A. Sinegub, 23 Apr 2005, MIMB 10517 (85); Sukhoi Liman, 46°19.9′N, 30°39.7′E, 0.3 m, sand, shell fragments, coll. I.A. Sinegub, 26 Jun 2001, MIMB 14119 (40); 0.3–13 m, mud, shell fragments, coll. I.A. Sinegub, 04 Sep 2002, MIMB 14120 (25). Gnele-Danube area: 45°52′N, 30°20′E, 14 m, muddy sand and shell fragments, coll. I.A. Sinegub, Aug 1992, MIMB 14116 (300+); 45°19′N, 29°49′E, 23 m, mud, coll. I.A. Sinegub, 25 Nov 2005, MIMB 14118 (4).

**Supplementary material**

GREECE, Aegean Sea, Sarinikos Gulf, near Psytalia Is., 37°55.42′N, 23°35.45′E, 65 m, sandy mud, coll. N. Simboura, 9 Feb 2012, MIMB 27201 (2).


**Adult morphology**

Up to 13 mm long and 1.1 mm wide for 60 chaetigers. Prostomium anteriorly incised to bifid, posteriorly extending to end of chaetiger 3 as a low caruncle. Two pairs of small black eyes and a prominent occipital antenna present on prostomium. Palps with a fine continuous black line along each side of frontal groove. Chaetiger 1 with short capillaries in neuropodia and small posterior lamellae in both rami; notochaetae absent. Chaetiger 5 with heavy falcate spines and companion chaetae arranged in a slightly curved oblique row; falcate spines each with a lateral tooth and a narrow thin subdistal longitudinal flange; companion chaetae each with a broom-like distal tip closely adhering to convex side of falcate spine; dorsal superior and ventral capillaries absent. Bidentate hooded hooks in neuropodia from chaetiger 7, not accompanied by capillaries. Branchiae...
from chaetiger 7 through most of body. Pygidium disc-like with dorsal gap.

**Remarks**

*Polydora cornuta* was originally described from Charleston Harbor, South Carolina, Atlantic USA by Bosc (1802) and later reported from estuaries and sea ports (mainly located in estuaries) all over the world (see Radashevsky, 2005). The first report of the species from the Black Sea was a misidentification. Pereyaslavtseva (1891) identified worms boring into gastropod shells occupied by hermit crabs off Sevastopol, Crimea, as *P. cornuta*, despite the fact that genuine *P. cornuta* is a tube-dwelling species unable to bore into calcareous substrata. The species was misidentified again when Losovskaya & Nesterova (1964) and Losovskaya (1969) reported large numbers of *Polydora ciliata limicola* Avenenkova, 1934 in Sukhoy Liman Lagoon in the Odessa area. Samples collected at that time no longer exist but discussion and joint examination of recent material by Galina V. Losovskaya and Vasily I. Radashevsky during VIR’s visit to Odessa in December 2005 helped to identify that material unambiguously as *P. cornuta* (see also Losovskaya, 2008: 360, 2011: 53). Adult worms were first recorded from the Odessa area in 1962 and, in 1963 population density reached 1155 ind.m⁻². In 2001, the population density of the species in Odessa Harbour reached 6600 ind.m⁻² in the benthic community and 9600 ind.m⁻² in the pier fouling communities (Losovskaya et al., 2004). Remarkably, no tube-dwelling (non-boring) *Polydora* worms were reported from the Black Sea before the 1960s, despite several studies on polychaetes in this region. It is likely therefore that Losovskaya & Nesterova (1964) reported the first arrival of *P. cornuta* in the Black Sea, at least in its northern part, which might actually have happened in the late 1950s.

As *P. ciliata*, *P. ciliata limicola*, *P. limicola*, and ultimately as *P. cornuta*, the species was reported from the Odessa area, Dnieper-Danube and adjacent regions in the north-western Black Sea (Losovskaya, 1976, 1977, 2008, 2011; Moroz, 1977, 1985, 1992; Sinegub, 2001, 2002a,b; Losovskaya & Sinegub, 2002; Losovskaya & Zolotarev, 2003; Losovskaya et al., 2004, 2005; Vorobyova et al., 2008; Bondarenko, 2009, 2011; Vorobyova & Bondarenko, 2009), Sevastopol in Crimea (Moroz, 1985; Kiseleva, 1988, 2004; Boltacheva & Lisitskaya, 2007), the Romanian coast (Surugiu & Manoleli, 1999; Surugiu, 2005a,b, 2006, 2012; Surugiu & Novac, 2007), and Tuapse Harbour, Russia (Selikonova, 2011). Kiseleva (1987) reported *P. ciliata limicola* for the first time from the Sea of Azov (Fig. 1A). Descriptions provided by these authors and our examination of the material reported in most of these ecological surveys (see Material section above) undoubtedly confirmed the presence of *P. cornuta*, as re-described by Blake & Maciolek (1987) and Radashevsky (2005). It is possible that earlier reports of *P. limicola* from the Romanian coast were also misidentifications of *P. cornuta* but the lack of the original material precludes verification (see Surugiu, 2005a).

Re-examination of old material from Izmir Bay on the Turkish coast of the Aegean Sea by Çinar et al. (2005b) revealed that *P. cornuta* was present in the region in 1986; sampling in this area in 2003 found a dense population of the species, up to 3170 ind.m⁻² (Çinar et al., 2005b). Populations of *P. cornuta* up to 170 ind.m⁻² were reported from Izmit Bay (Dağlı & Ergen, 2008) and 3390 ind.m⁻² from Kalamış Bay (Karhan et al., 2008), both in the Sea of Marmara; a dense population of the species up to 4340 ind.m⁻² was reported from the Golden Horn Estuary in the Bosphorus Straight (Çinar et al., 2009). In the Dniester estuary, the population density of the species reached 54904 ind.m⁻² (Vorobyova & Bondarenko, 2009), and in severely polluted areas on the Romanian coast it reached 95000 ind.m⁻² while the biomass attained 157.4 g.m⁻² (Surugiu, 2005b). In the Mediterranean Sea, *P. cornuta* was reported from Valencia Harbour in Spain (Tena et al., 1991), and Elefsis Bay on the Greek coast of the Aegean Sea (Simboura et al., 2008) (Fig. 1A).

*Polydora cornuta* is an opportunistic species tolerant of wide ranges of salinity and temperature, and characterized by early maturation, iterative breeding in water temperatures above 13°C, high larval production, ability to colonize disturbed and polluted substrates and to establish high density populations in a short time (Dauer et al., 1981; Zajac, 1991a,b; Radashevsky, 2005). It is being widely distributed by shipping and aquaculture. Planktrophic larvae of the species survive transportation in ballast water, and adults may form dense settlements through fouling of ship hulls (Radashevsky, in preparation). The routes of the species in the Mediterranean, Black Sea and the Sea of Azov remain uncertain. The limited number of recent reports from the Mediterranean Sea (in contrast to many reports from the Black Sea) may be due to the limited number of taxonomic studies rather than a limited distribution of the species in the region. It is possible that *P. cornuta* was either first introduced by ocean-going vessels to the north-western part of the Black Sea and then transported to the eastern Mediterranean or that there were several independent introductions into the Mediterranean Sea and Black Sea. The native distribution of the species is unknown and may no longer be possible to determine because of numerous transports by human activities.

**Distribution**

Widespread, mainly in estuaries and sea ports.
**Streblospio gynobranchiata** Rice & Levin, 1998


**Adult morphology**

Up to 12 mm long and 0.5 mm wide for 55 chaetigers. Prostomium anteriorly rounded, posteriorly extending to chaetiger 1. Peristomium forming a prominent hood enveloping prostomium laterally and ventrally (Fig. 2A–C). Two pairs of small red eyes arranged trapezoidally. Occipital antenna absent on prostomium. Palps as long as 5–10 chaetigers, with up to 20 small transverse bands/spots of dark pigment. Short median papilla present on dorsal side of chaetiger 1. Chaetiger 1 with short capillaries in both rami and small postchaetal lamellae in neuropodia; notopodial lamellae absent. Notopodial postchaetal lamellae of chaetiger 2 interconnected by a prominent transverse membrane or collar, forming a characteristic pouch open anteriorly; median part of collar straight to slightly concave (Fig. 2C). Sabre chaetae

**Material**

RUSSIA, north-eastern Black Sea: Novorossiysk Harbour, Novorossiysk Bay, 44º43.55′N, 37º47.1′E, 10 m, mud, 1 Aug 2007, MIMB 27180 (3); 31 Jul 2008, MIMB 27181 (70); 3 Sep 2010, MIMB 27182 (75). Bolshoi Utrish, Snake Lake Liman, 44º45.92′N, 37º23.24′E, 3 m, mud, 28 Aug 2011, MIMB 27183 (7); 15 Nov 2011, MIMB 27184 (18).

UKRAINE, north-western Black Sea, Odessa area, Sukhoi Liman, 46º19.9′N, 30º39.7′E, 0.3–13 m, mud, shell fragments, coll. I.A. Sinegub, 04 Sep 2002, MIMB 14124 (10).
in neuropodia from chaetiger 7. Hooks in neuropodia from chaetiger 7, up to 9 in a series, accompanied by 1–3 alternating capillaries and 1–2 inferior sabre chaetae. Hooks multidentate, with 3–5 pairs of small upper teeth arranged in two vertical rows above main fang. Single pair of branchiae on chaetiger 1 situated just posterior to pair of palps; branchiae apinate, flattened, with undulating lateral edges, almost equal in length to palps and appearing similar to them (Fig. 2A–C). Finger-like extensions of lateral body wall up to 350 µm long and 50 µm in diameter in mature females from chaetigers 16–19 to chaetigers 22–33; extensions oriented dorsally and posteriorly and containing mature oocytes (Figs 2E–G, 3C, D). Pygidium small, rounded, with one pair of very short ventral knobs (Fig. 3D). In males, sperm from chaetiger 7 to chaetigers 20–36. In females, oocytes from chaetiger 8 to chaetigers 25–33. Oocytes up to 85 µm in diameter, with germinal vesicle 30 µm, and single nucleolus 10 µm in diameter; up to about 10 large vitellogenic oocytes present in each ovary and 20 oocytes in each fertile segment (Figs 2D–F, 3C, D). Unpaired intraepithelial seminal receptacles present in females on ventral side of 2–3 chaetigers, from chaetigers 13–14 to chaetigers 14–15; receptacles oval to fusiform, up to 110 µm long and 30 µm wide, oriented longitudinally between ventral nerve cords (Fig. 3A, B).

Habitat
In Novorossiysk Harbour, adults of *S. gynobranchiata* usually occur in the estuary of River Tsemess, with water salinity at 12.9 ‰. They inhabit the upper layer of muddy sediments containing hydrogen sulphide. The population density of the species reached 980 ind.m$^{-2}$ in 2001, 9000 ind.m$^{-2}$ in 2007 and 10000 ind.m$^{-2}$ in 2008.

Remarks
*Streblospio gynobranchiata* was originally described from Tampa Bay, Florida, USA by Rice & Levin (1998) and later reported from the Aegean Sea, Turkey (Çinar et al., 2005 a,b, 2008, 2012a; Dağlı et al., 2011), Crimea in the northern Black Sea (Boltachova, 2008), the southern part of the Caspian Sea (Taheri et al., 2008; Taheri & Foshtomi, 2011), and the Golden Horn Estuary, Bosphorus Strait (Çinar et al., 2009) (Fig. 1A). In all localities, worms occurred on muddy substrata near to and within the port reaches, reaching maximum density of 60480 ind.m$^{-2}$ in Alsancak Harbour (Izmir Bay of the Aegean Sea) (Çinar et al., 2009). Because the species was not found in earlier biological surveys conducted in these areas, it is classified as an alien, most likely introduced with ballast waters.

Worms collected within the framework of this study fit all the features of *S. gynobranchiata* (see Rice & Levin, 1998) and are characterized by the presence of a single pair of branchiae situated close to and appearing similar to palps, a dorsal pouch on chaetiger 2, lateral extensions of the body wall, and hooks in neuropodia consistently beginning from chaetiger 7. Remarkably, the body wall extensions are present only in mature females but absent in immature individuals and males.

Of the three currently recognized *Streblospio* species (see Mahon et al., 2009), *S. benedicti* Webster, 1879 and *S. gynobranchiata* have been considered as native to the Americas, while *S. shrubsolii* (Buchanan, 1890) seems to be native to European waters. Adults of *S. gynobranchiata* are similar to *S. benedicti* in having neuropodial hooks starting from chaetiger 7, and both differ from *S. shrubsolii*, which have hooks starting from chaetigers 8–10. Males of *S. gynobranchiata* and *S. benedicti* appear quite similar to each other, but females differ in having lateral body wall extensions in *S. gynobranchiata* and dorsal brooding pouches in *S. benedicti*. As one of the most common polychaete species on both coasts of North America, and also due to poecilogonous ability to produce planktotrophic and lecithotrophic larvae, *S. benedicti* became a subject of numerous studies on ecology and reproductive biology (e.g. Levin & Huggett, 1991; Mccain, 2008; Gibson et al., 2010). The other two species have been less thoroughly studied and morphological differences between them have remained uncertain.

Distribution
Atlantic coast of North and South America from Florida, U.S.A. to Paraná, Brazil; Mediterranean, Black and Caspian Seas.

Conclusion
Alterations (addition - invasion, deletion - extinction, and altered population dynamics) to the biodiversity of terrestrial, freshwater and marine communities are the expected and inexorable consequences of human activity currently seen throughout the world (Carlton, 2009). However, recognizing these alterations is not an easy task except where large, well-known species are of concern. Although alien, non-indigenous species have become subjects of study in many countries in recent years, these studies are still mainly based on morphological examination and comparison of fixed specimens. When the biogeographic origin of a species and its morphological variability are unknown, identification of specimens from remote places often remains ambiguous and uncertain.

Spioid polychaetes demonstrate an amazing diversity of morphological, ecological and reproductive characteristics and show significant variability (both ontogenetic and individual) in important taxonomic characters (Radashevsky, 2012). The geographic morphological
variability of spionids is poorly studied but should be considered when making identifications. Molecular techniques, currently under rapid development, are certainly needed not only for the identification of species but also for recognition of their biogeographic origins and possible routes of distribution. Molecular comparisons of spionids from distant places will provide closer insight into their taxonomy and distribution.

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