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Rapid Communication

First record of the non-indigenous *Melita nitida* Smith, 1873 (Crustacea: Amphipoda: Melitidae) in the Bay of Biscay (NE Atlantic)

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Abstract

The non-native amphipod *Melita nitida* Smith, 1873 was collected between 2013 and 2016 in Arcachon Bay, Hossegor Lake and the Gironde Estuary (SW France) in intertidal oyster reefs and under stones. This species, native to the Atlantic coast of North America, is considered as a non-indigenous species on the Pacific coast of North America. Recently, the presence of *M. nitida* was reported in Europe, both in The Netherlands and in Germany. This species may have been accidentally introduced to Arcachon Bay with oyster transfers. An identification key for species belonging to the genus *Melita* and closely related species in European waters is also provided.

Key words: non-indigenous species, Arcachon Bay, Hossegor Lake, Gironde Estuary, oyster transfer

Introduction

The genus *Melita* Leach, 1814 is diverse and includes 78 valid species according to the World Register of Marine Species (Lowry 2010a; *Melita mirzajanii* Krapp-Schickel and Sket, 2015 not yet included in this database). Thirteen species are known from European waters (Lowry 2010b): *M. abyssorum* Stephensen, 1944, *M. bulla* Karaman, 1978, *M. coroninii* Heller, 1866, *M. dentata* (Krøyer, 1842), *M. formosa* Murdoch, 1866, *M. hergensis* Reid, 1939, *M. nitida* Smith, 1873, *M. pallida* G.O. Sars, 1879, *M. palmata* (Montagu, 1804), *M. reidi* Hamond, 1965, *M. richardi* Chevreux, 1900, *M. valesi* Karaman, 1955 and *M. virgula* Krapp-Schickel, Ruffo and Schiecke, 1994. Only three of the European species are known to lack teeth on pleosome and urosome: *M. bulla*, *M. nitida* and *M. valesi*, as well as another species previously classified in the genus *Melita*: *Allomelita pellucida* (Sars, 1882).

Melita nitida was originally described from New England (NE America) by Smith in 1873. According to Bousfield (1973), this species occurs from the southwestern Gulf of St. Lawrence (Canada) to the Yucatan Peninsula (Mexico) in the NW Atlantic. Outside its native range, Chapman (1988) recorded *M. nitida* in the NE Pacific from British Columbia to California and considered records from Ecuador, Panama, and Costa Rica (Shoemaker 1935) as dubious —possibly representing an undescribed species. In European waters, *M. nitida* has been observed since 1998 in The Netherlands (Faasse and van Moorsel 2003) and 2010 in Germany (Reichert and Beermann 2011). Here we report the first record of *M. nitida* in three localities of the French Atlantic coast: Arcachon Bay, Hossegor Lake and the Gironde Estuary. An identification key for species belonging to the genus *Melita* in European waters is also provided (Table 1).

Material and methods

Study areas

The San Arcachon Bay (Figure 1) is a 180 km² macrotidal coastal lagoon, connected to the Atlantic Ocean by a narrow channel and receives freshwater inputs in its south-eastern part (Leyre River). This lagoon is characterized by large intertidal flats (115 km²)

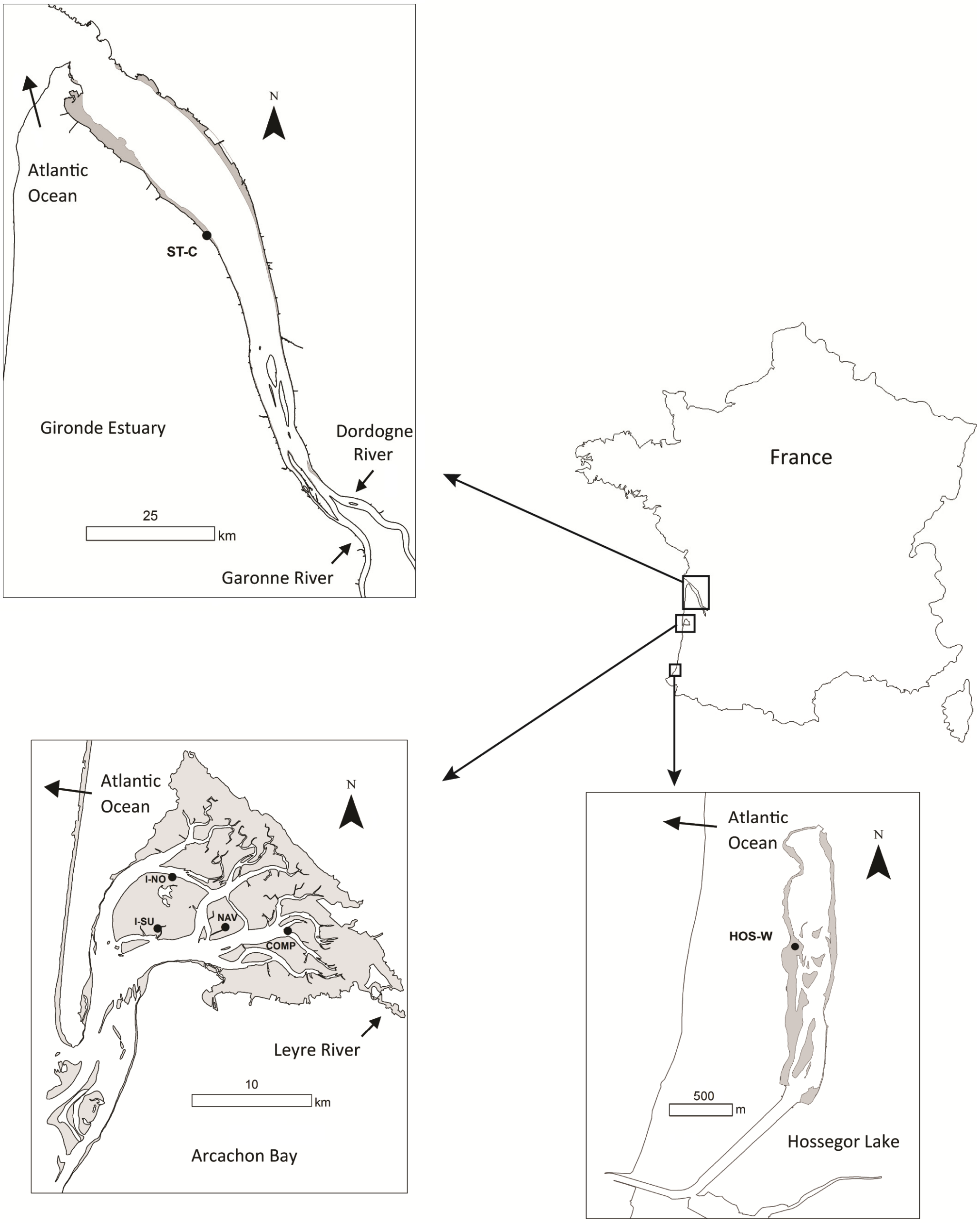

Figure 1. Sampling stations for *Melita nitida* in Arcachon Bay, Hossegor Lake and the Gironde Estuary (see code legend in Supplementary material Table S1). Shaded areas indicate intertidal zones.

covered by *Zostera noltei* Hornemann, 1832 seagrass bed (70 km²) (Plus et al. 2010). The lower parts of tidal flats are mainly occupied by oyster farms (10 km²) [Pacific oysters, *Crassostrea gigas* (Thunberg, 1793)]. In the inner lagoon, tidal channels represent 71 km², with 1.02 km² occupied by eelgrass beds [*Zostera marina* Linnaeus, 1753] (Plus et al. 2010). At the sampling stations, salinity ranges annually between 24 and 35.

Hossegor Lake (Figure 1) is a small (0.9 km²), shallow, marine water body connected to the Atlantic Ocean by a very narrow channel. Intertidal flats represent 0.28 km², and *Z. noltei* covers 0.012 km² (Trut et al. 2014). The lake has been highly modified

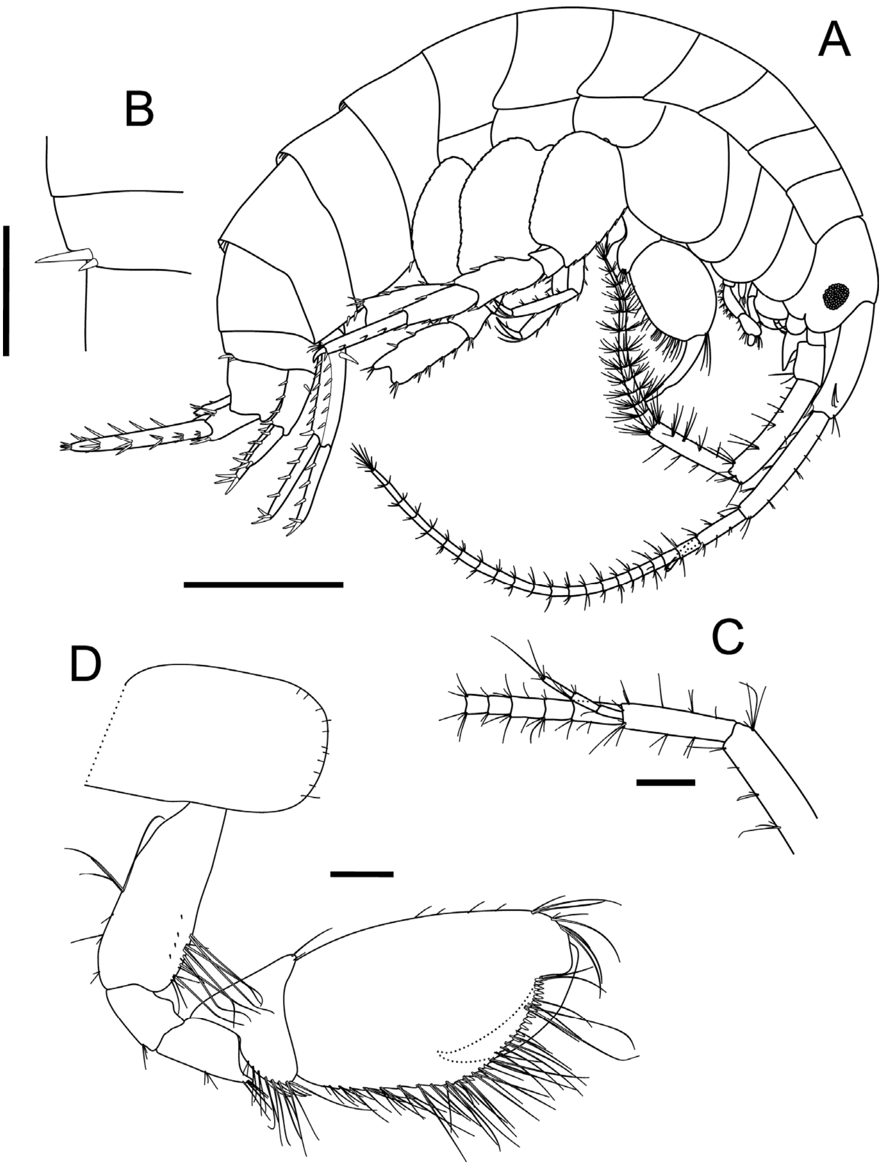

Figure 2. *Melita nitida* Smith, 1873 from Ile Nord, Arcachon Bay, November 2014. Male, BL = 6.95 mm, A) lateral view, B) urosomes 1–3. Male, BL = 6.03 mm, C) right antenna 1, inner face of peduncle article 2 (in part), article 3, accessory flagellum and flagellum (in part), D) right gnathopod 2, outer face. Scale bars: A: 1 mm; B, C, D: 0.2 mm.

by human activities (embanking, oyster farming, dredging). The maximum water depth is 4 m and the salinity ranges between 21 and 35 (Auby et al. 2015).

The Gironde Estuary is the largest SW European estuary, with a total haline area of 625 km²; intertidal mudflats occupy about 8% of this area. Water depth in the navigation channel ranges between 6 m at the confluence of the Garonne and Dordogne rivers to30 m at the mouth of the estuary. The sampling station was located on the western bank in the middle estuary (Figure 1) where water salinity and temperature vary annually between 0.5 and 17.3 (mean 8.3) and 7 and 26°C (mean 18.6°C), respectively. Although oyster farming has been prohibited since 1996 due to cadmium pollution, wild oysters occur on hard intertidal substrates along the haline part of the estuary. Unlike the previous two locations, the Gironde Estuary receives international maritime traffic, with 8.2–9.0 million tonnes of freight circulating annually (http://www.bordeaux-port.fr/fr/chiffres-clés).

Material examined

Melita nitida was collected at six intertidal stations: four in Arcachon Bay, one in Hossegor Lake, and one in the Gironde Estuary (Figure 1, Supplementary material Table S1). Specimens were collected in oyster reefs, under stones, and among oysters on wooden pillars. The specimens taken back to the laboratory were examined under a Nikon SMZ 1500 stereomicroscope and a Nikon Eclipse E400 microscope with up to 112.5 and 400 × magnifications (and transmitted light) respectively, and photographed with a Nikon DS-Fi2 camera. Illustrations were made from photographic images using Inkscape software (v.0.48). Body length (BL) was measured with NIS-Elements Analysis software (Nikon Instruments Inc., Melville, NY, USA) from the anterior margin of head to the posterior end of telson. Six specimens were deposited in the Muséum National d'Histoire Naturelle (MNHN, Paris) (Table S1).

Results

We collected 76 specimens from the three locations, mainly in oyster reefs: 42 males, 24 brooding females, and 10 females with oostegites. These specimens were collected between January 2013 and November 2014 in Arcachon Bay, in July 2014 in Hossegor Lake, and in February 2016 in the Gironde Estuary (see Table S1). Most specimens (n=64) were collected in Arcachon Bay, which may be partially attributed to a higher sampling effort in this area.

Description (Figure 2)

Based on adult male, 6.95 mm (BL), Arcachon Bay (SW France), station 'Ile Nord', November 15, 2014.

Body smooth, urosome segment 2 with 3 spines. *Head* with eyes normally developed, ovoid; antennal sinus present. *Antenna 1* longer than antenna 2; peduncle article 1 slightly shorter than article 2, with two proximal spines on inner face and one posterodistal spine; article 3 short; flagellum with 22 articles; accessory flagellum with 4 articles (terminal articles minute). *Antenna 2* peduncle article 2 cone gland not reaching article 3, with an anterodistal spine; article 3 short, with one dorsal and two median spines on inner face; article 4 longer than article 5, with a row of 7 spines on median inner face; article 5 with a row of 6 clusters of setae – spines on median inner face (variability on clusters setation in the same specimen: 0–2 spines with 1–4 setae per cluster); flagellum with 12 articles, with many dense setae. *Mandibular* palp article 1 short, without setae; article 2 shorter than article 3, with 3 lateral and 4 median setae; article 3 with 6 lateral and 3 distal setae. *Gnathopod 1* smaller than gnathopod 2, subchelate; coxa with short setae on ventral margin; basis slender, with long setae on distal half of anterior margin and a cluster of 4 long setae on the middle of the posterior margin; merus covered with many short setae on posterior to inner median margin; carpus longer than propodus; propodus longer than broad, palm with row of slender spines. *Gnathopod 2* subchelate; coxa and basis similar to gnathopod 1; propodus longer than broad, inner face densely setose with a weak excavation defined by a row of short spines and setae in which dactylus rests, palm regularly convex. *Pereopods 3* and *4* slender, poorly setose; dactylus with a pappose seta on posterior margin. *Pereopods 5–7* posterior margin of basis serrated, distal part straight to slightly concave with a small posterodistal lobe. *Epimera 1–3* subquadrate with a small tooth on posterodistal corner, very slightly serrated and with short setae on posterior margin; epimeron 1 ventral margin smooth; epimera 2–3 proximal part of ventral margin with 1 and 2 spines, respectively. *Uropod 1* peduncle with one basofacial spine, another stronger one distally, and a row of lateral and marginal spines. *Uropod 2* peduncle subequal in length to rami. *Uropod 3* much exceeding tip of uropod 1; peduncle shorter than outer ramus; inner ramus scale-like; outer ramus 1-articulate, marginally and distally spinose. *Telson* cleft near to the base with 1 subapical spine on outer margin, 1 medial and 2 subapical spines on median margin of each lobe; top of the lobes subacute, without setae or spines.

Female (sexually dimorphic characters based on brooding female, 6.36 mm (BL), 23 eggs in the marsupium, Arcachon Bay (SW France), station "Ile Sud", January 14, 2013).

Antenna 2 peduncle articles 4 and 5 without spines on inner face, flagellum without dense setae. *Gnathopod 2* less powerful and setose than in male; propodus without excavation, proximal end of palm limited by a strong spine on inner face. *Coxa 6* with anterodistal cusp.

Variability

Melita nitida shows variability in ornamentation on urosome segment 2, which is size-dependent (Reichert and Beermann 2011). This was confirmed by observations of a significant positive correlation between the number of spines (range: 1–5) and the length of the specimens (BL range: 2.4–8.64 mm) (non-parametric Spearman, r = 0.68, p<0.01). There was also a positive correlation between the length of the organism and the number of articles of the accessory flagellum (range: 3–5) (non-parametric Spearman, r = 0.73, $p<0.05$), but there was no significant difference in the number of spines between genders (Kruskall-Wallis test, $p>0.05$). In some cases, there was no right-left symmetry in terms of number of urosome 2 spines, in number of accessory flagellum articles, or in the presence / absence of telson medial spine on median lobe margin.

Considering all available information (i.e. Faasse and van Moorsel 2003; Reichert and Beermann 2011, current survey), the main characters to distinguish *M. nitida* from other European species of the "*Melita*-group" are: (1) the absence of dorsal teeth on pleosome and urosome; (2) eyes normally developed; (3) antennal sinus present; (4) antenna 1 accessory flagellum with at least 3 articles; (5) male antenna 2 flagellum with dense setae; and (6) only urosome segment 2 with dorsal spines.

Discussion

Melita nitida was redescribed by Mills in 1964 because the original description was insufficient to distinguish it from other *Melita* species. Krapp-Schickel and Sket (2015) created four groups of melitid species, based on some morphological characters (pleosome and urosome teeth and uropod 3 outer ramus one or two-articulate) but not on phylogenetic relationships. *M. nitida* belongs to *Melita*-Group B1: pleosome and urosome lacking dorsal teeth (i.e. non-articulated acutely ending cuticular protuberances), urosome beset with articulated cuspidate setae; U3 outer ramus with one article. This group contains 26 species (25 *Melita* species and 1 *Allomelita* species), including two species closely related to *M. nitida*: *M. elongata* Sheridan, 1980 and *M. setiflagella* Yamato, 1988. *M. elongata* is distinguished from *M. nitida* and *M. setiflagella* by urosome 2 ornamentation: 2 cuspidate setae on each side dorsolaterally for *M. elongata* (specimen 5 mm) versus 3 or more for *M. nitida* and *M. setiflagella* (see Krapp-Schickel and Sket 2015). *M. elongata* can also be distinguished by its accessory flagellum, which is composed of 1–2 short articles (Sheridan 1979), whereas *M. nitida* and *M. setiflagella* have up to 3 articles. *M. nitida* is very closely related, morphologically, to *M. setiflagella* from Japan, but differs by the absence of a notch at inferior antenna 1 corner of head and the setation of antenna 2 peduncle article 5. Krapp-Schickel and Sket (2015) consider *M. nitida* and *M. setiflagella* as extremely similar, morphologically, and some authors (Jarrett and Bousfield 1996; Faasse and van Moorsel 2003; Reichert and Beermann 2011) even consider both species as synonyms.

Within the 25 *Melita* species belonging to *Melita*-Group B1, only four have been recorded from European waters: *M. bulla*, *M. nitida, M. valesi,* and *Allomelita pellucida* (previously *Melita pellucida* Sars, 1883). According to Krapp-Schickel and Sket (2015), *M. nitida* differs from these other European species by the absence of cuspidate setae on urosome 1 (vs. 1 pair of cuspidate setae for *A. pellucida*), formation of normally developed eyes (vs. weakly developed or absent for *M. valesi*), and the gnathopod 2 male propodus being rectangular with a palmar corner >120° (vs. gnathopod 2 male propodus being subrectangular with a palmar corner ≤ 120° for *M. bulla*). Karaman (1982) described *M. bulla* and *M. valesi* as endemic to the Mediterranean Sea. *Allomelita pellucida* is the only native Atlantic species without teeth on urosome and pleosome. This species differs from *M. nitida* (in addition to ornamentation of urosome 1) by accessory flagellum one-articulated and setation of antenna 2 (Chevreux and Fage 1925; Lincoln 1979). Only two *Melita* species, namely *M. hergensis* and *M. palmata*, have been recorded so far in the Bay of Biscay (Bachelet et al. 2003). The present study thus reports the first records of *M. nitida* in this area. However, *M. nitida* may have been previously present, but its low abundance level and few studies in particular habitats of this species could explain why it has not been reported before.

In our study, *Melita nitida* was mainly found in oyster reefs, which is in agreement with previous authors who indicated it as commonly associated with oysters (Watling and Maurer 1972) or occurring among intertidal rocks and seaweeds (Paulmier 1905, Kunkel 1918). Our specimens were found in polyhaline waters in Arcachon Bay and Hossegor Lake, which is within the optimal salinity range recorded by Sheridan (1979), and in mesohaline waters in the Gironde Estuary. *Melita nitida* is known to occur within a wide temperature (0 to 32°C) and salinity (0 to 35) range (Bousfield 1973; Sheridan 1979; Chapman 1988; Faasse and van Moorsel 2003; Reichert and Beermann 2011) and thus has a high invasive potential.

The numerous introductions of non-indigenous species in Arcachon Bay are well documented. Ballast-water is an unlikely vector of introduction since there is no large harbour or shipping traffic in Arcachon Bay and Hossegor Lake. The most likely vectors are fouling on recreational boats and transfers of oysters (*Crassostrea gigas*). Indeed, local oyster farmers often transfer their productions among rearing areas in France (both on the Atlantic and Mediterranean coasts) (Goulletquer et al. 2002) or even in other European countries (B. Simonnet, Service

Table 1. Identification key to adults of *Melita* species from European waters (according to the biogeographical limits of the European Register of Marine Species). *Allomelita pellucida* and *Abludomelita* species are included in this key for their great resemblances to *Melita* species.

01.	Uropod 3 outer ramus 2-articulate. Maxilla 2 inner plate with medial row of setae	2 (*Abludomelita*)
-	Uropod 3 outer ramus 1-articulate. Maxilla 2 inner plate without medial row of setae	4
02.	Pleosome segments 1–3 distal margin with 3 teeth	*A. gladiosa* (Bate, 1862)
-	Pleosome segment 1 distal margin smooth or with 1 tooth, pleosome segment 3 distal margin with 0–1 tooth	3
03.	Epimeral plate 3 posterior margin smooth	*A. obtusata* (Montagu, 1813)
-	Epimeral plate 3 posterior margin serrate	*A. aculeata* (Chevreux, 1911)
04.	Pleosome with teeth	5
-	Pleosome without teeth	9
05.	Pleosome segment 1 lacking teeth	*Melita formosa* Murdoch, 1866
-	Pleosome segment 1 with 1 or more teeth	6
06.	Eyes weakly developed or absent	7
-	Eyes normally developed	8
07.	Pleosome segments 1–3 with 1 dorsal tooth	*M. richardi* Chevreux, 1900
-	Pleosome segments 1–3 with at least 2 dorsal teeth on each side	*M. pallida* G.O. Sars, 1879
08.	Pleosome segments 1–3 with several small teeth; antenna 1 accessory flagellum usually 4-articulate	*M. dentata* (Krøyer, 1842)
-	Pleosome segments 1–3 with 1 dorsal tooth; antenna 1 accessory flagellum 2-articulate (the last article very small)	*M. virgula* Krapp-Schickel, Ruffo and Schiecke, 1994
09.	Urosome with dorsal teeth	10
-	Urosome without dorsal teeth	14
10.	Urosome segment 1 with one dorsal tooth	11
-	Urosome segment 1 without dorsal tooth	*M. coroninii* Heller, 1866
11.	Eyes weakly developed or absent	12
-	Eyes normally developed	13
12.	Antenna 1 accessory flagellum 2-articulate; urosome segment 3 with 1 small lateral tooth on each side	*M. reidi* Hamond, 1965
-	Antenna 1 accessory flagellum 5-articulate; urosome segment 3 without teeth	*M. abyssorum* Stephensen, 1944
13.	Antennal sinus absent; telson without distal spines	*M. hergensis* Reid, 1939
-	Antennal sinus present; telson with distal spines	*M. palmata* (Montagu, 1804)
14.	Urosome segments 1 and 2 with dorsal spines, telson cleft to three-quarters length	*Allomelita pellucida* (Sars, 1882)
-	Urosome segment 1 always totally naked, telson cleft nearly to base	15
15.	Eyes weakly developed (2–6 ommatidia) or absent	*M. valesi* Karaman, 1955
-	Eyes normally developed	16
16.	Antennal sinus absent; male antenna 2 flagellum not very setose	*M. bulla* Karaman, 1978
-	Antennal sinus present; male antenna 2 flagellum with many dense setae	*M. nitida* Smith, 1873

Mer et Littoral, Direction Départementale des Territoires et de la Mer de la Gironde, pers. comm.) to increase bivalve growth and reproduction. The transfer of *C. gigas* among regions has caused the worldwide introduction and spread of numerous alien species (e.g., Goulletquer et al. 2002; Wolff and Reise 2002; Ruesink et al. 2005; Haupt et al. 2010; Mineur et al. 2014). Several non-indigenous species, probably introduced with oyster transfers, were recently found in Arcachon Bay: the bivalve *Arcuatula senhousia* (Benson in Cantor, 1842) (Bachelet et al. 2009), the isopod *Paranthura japonica* Richardson, 1909 (Lavesque et al. 2013), and the amphipods *Grandidierella japonica* Stephensen, 1938 (Lavesque et al. 2014), *Aoroides curvipes* Ariyama, 2004, *Aoroides longimerus* Ren and Zheng, 1996 and *Aoroides semicurvatus* Ariyama, 2004 (Gouillieux et al. 2015). Since no published records are currently known for *M. nitida* between The Netherlands and the southwestern French coast, the introduction of *M. nitida* in Arcachon Bay and Hossegor Lake is likely due to oyster transfers from British Columbia (Pacific Canada) or from Northern European sites. *M. nitida* may be present in other oyster farming sites, but this has not yet been documented. Concerning the Gironde Estuary, there are currently no more oyster farms present due to cadmium pollution, but some patches of natural oyster beds are still present. However, maritime traffic is well developed in the Gironde Estuary. Some species, such as the shrimp *Palaemon macrodactylus* Rathbun, 1902 and the copepod *Acartia tonsa* Dana, 1849, were introduced in the Gironde Estuary through ballast ballast waters (Beguer et al. 2007; David et al. 2007), and *M. nitida* may have also been introduced to this location via ballast water. Molecular studies could be helpful in determining the source of different *M. nitida* populations in these three water bodies.

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Supplementary material

The following supplementary material is available for this article:

Table S1. Records of *Melita nitida* in Arcachon Bay, Hossegor Lake and the Gironde Estuary.

This material is available as part of online article from:

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