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Rapid Communication**First records of *Aoroides longimerus* Ren and Zheng, 1996, and *A. semicurvatus* Ariyama, 2004 (Crustacea, Amphipoda), in the English Channel, France**

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OPEN ACCESS**Abstract**

Two Pacific amphipod species, *Aoroides longimerus* (94 individuals) and *A. semicurvatus* (three individuals), were recorded for the first time in 2019 in the English Channel. *Aoroides longimerus* was found in four sites along the Normandy coast (Granville, Cherbourg, Saint-Vaast-La-Hougue, and Le Havre) while *A. semicurvatus* was found only in two sites (Granville and Saint-Vaast-La-Hougue). Because both species were recorded (2009–2014) from the French Atlantic coast, they have probably been introduced to the English Channel through the accumulated fouling of the hulls of ships used primarily for recreational purposes or by transfer of the Japanese oyster *Crassostrea gigas* between oyster farms along the French oyster production intertidal zones.

Key words: introduced species, Le Havre harbour, non-native species, fouling, Bay of Seine

Introduction

Among the 151 non-native species (NNS) recorded in Normandy (French coast of the English Channel) at the end of 2018, the crustaceans, mainly barnacles, amphipods and decapods with 34 species, was the richest zoological group (Baffreau et al. 2018; Pezy et al. 2020). Among these crustaceans, seven were amphipods: *Caprella mutica* Shurin, 1935, *Cryptorchestia cavimana* (Heller, 1865), *Dikerogammarus villosus* (Sowinsky, 1894), *Grandidierella japonica* (Stephensen, 1938), *Monocorophium acherusicum* (Costa, 1853), *Monocorophium sextonae* (Crawford, 1937), and *Platorchestia platensis* (Krøyer, 1845). A further eight more species had the potential to be recorded in the future in Normandy waters, viz., *Aoroides curvipes* Ariyama, 2004, *Aoroides longimerus* Ren and Zheng, 1996, *Aoroides semicurvatus* Ariyama, 2004, *Chelicorophium curvispinum* (G.O. Sars, 1895), *Gammarus tigrinus* (Sexton, 1939), *Melita nitida* (Smith, 1873), *Monocorophium uenoi* (Stephensen, 1932), and *Ptilohyale littoralis* (Stimpson, 1853), as these have been recorded in neighbouring zones in

the eastern part of the English Channel and the southern part of the North Sea, Brittany or the Bay of Biscay (Jourde et al. 2013; Lavesque et al. 2014; Spilmont et al. 2016; Gouillieux et al. 2015, 2016; Gouillieux 2017; Fouquier et al. 2018; Gouillieux and Massé 2019).

Two pathways of NNS introduction in Normandy have been proposed. The first is maritime transport; in particular, Le Havre, Rouen, Dunkirk, Cherbourg and Calais harbours represent 15% of the maritime traffic of the global volume of maritime shipping in the English Channel (Bahé 2008). Le Havre harbour is the first place for the records of NNS in the English Channel (Ruellet and Breton 2012; Breton 2014; Baffreau et al. 2018; Pezy et al. 2020). Another pathway is aquaculture, mainly of the Japanese oyster *Crassostrea gigas* (Thunberg, 1793) and its transfer between the Atlantic and English Channel oyster centers of production (Baffreau et al. 2018). Since our last NNS inventory in Normandy (end of 2018), we continued to survey the macrofauna of the intertidal zone plus those of marinas and ports along the Normandy coast. Newly recorded NNS that were previously categorised as potential introduced species in Baffreau et al. (2018) have been discovered, including the isopod *Paranthura japonica* Richardson, 1909 (Pezy et al. 2020) and two species of the amphipod genus *Aoroides* Walker, 1898 (present paper). Gouillieux et al. (2015) first reported this Pacific amphipod genus in European waters with three species recorded in the southern part of the Bay of Biscay and Bay of Brest along the French Atlantic waters in 2009–2014: *Aoroides semicurvatus* and *A. curvipes*, in the Arcachon Bay and Hossegor Lake; and *A. longimerus*, in the Arcachon and Brest Bays. Male specimens had been reported from oyster reefs, floating pontoons in marinas, *Zostera* meadows and slipper limpet beds (Gouillieux et al. 2015). Two of these three species originated in Japan (Ariyama 2004), while *A. longimerus* originated from China (Ren and Zheng 1996). In its native range, *A. longimerus* occurs in subtidal hard substratum with biogenic structures, among algae and sessile organisms. *A. semicurvatus* was reported from the lower intertidal zone, under stones, rarely among algae (Ariyama 2004). The third species *A. curvipes* was found mainly on sandy mud bottom (Ariyama 2004). Recently during the 2017 autumn, several individuals of the species *A. semicurvatus* were reported from the Oosterschelde, Netherlands (Faasse et al. 2018). Faasse et al. (2018) suggested that shellfish imports, or, less likely, transport with recreational vessels, could be the introduction vector.

Harbours and marinas are known to be the receptacle of numerous NNS including vagile species and among them amphipods (Bishop et al. 2014; López-Legentil et al. 2015; Ros et al. 2013b). Some amphipods possess very high ability to colonise and then disperse from a colonised area after their introduction, such as *Caprella mutica* (Breton 2004; Ashton et al. 2010; Almón et al. 2014) or *Caprella scaura* Templeton, 1936 (Cabezas et al. 2014). They colonise dykes, pontoons, and the hulls of the boats in fouling

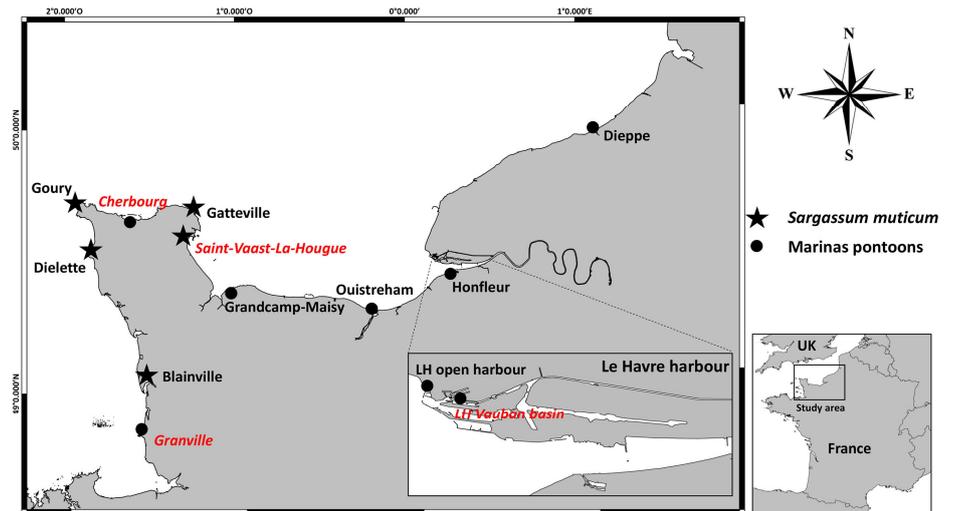


Figure 1. Sampling sites of the eight marinas and five *Sargassum muticum* zones visited in Normandy in 2019 during the framework of the ENBIMANOR project. LH: Le Havre. In italics and red, sites where the *Aoroides* species had been recorded.

communities, which ensures establishment success of these NNS (Ros et al. 2013a, 2016; Zabin et al. 2014). Vessels and biofouling have been sources of numerous introductions of amphipod NNS in oceans worldwide (Hewitt et al. 2009; Kaluza et al. 2010; Seebens et al. 2013. Marchini and Cardeccia 2017); this includes recreational boating (Cohen et al. 2005; Darbyson et al. 2009; Davidson et al. 2010; Clarke Murray et al. 2011; Zabin et al. 2014).

The aim of this paper is to report the recent records of *Aoroides longimerus* and *A. semicurvatus* in the Normandy zone of the English Channel.

Materials and methods

In the framework of the ENBIMANOR project (ENrichissement de la Biodiversité MARine en NORmandie) in Normandy, field surveys were planned during the period 2019–2021 to prospect both the macrofauna living in coastal marinas and ports, and along the intertidal zone (Figure 1). In eight marinas from Granville in the west to Dieppe in the east, samples of the fauna were collected by scraping the hard substratum that had formed on the floating pontoons in winter 2019 from 24 January to 1 March. Five replicates were collected from each site, for a total sampling area covering approximately 0.3 m². Detached material was collected in a Working Party 2 (WP2) net (200 µm) in order to avoid loss of scraping organisms.

Meanwhile, distribution of the macrofauna associated with a brown alga *Sargassum muticum* (Yendo) Fensholt, 1955, was also studied from 4 to 16 April 2019 along the Cotentin Peninsula in five zones (Figure 1) where abundant populations of this non-native invasive algal species were present (Baffreau et al. 2018). The sampling method consisted of using a suprabenthic sledge with a single 0.18 m² box (0.6 × 0.3 m) to filter the water at approximately 0.1–0.40 m above the sea bottom in rock pools with water at low tide in dense *S. muticum* populations. The box was equipped

with a WP2 zooplankton net (500 μm mesh size), the sledge was operated using three transects of 5 m each corresponding to a distinct sampling surface of 3 m^2 and a total surface of 9 m^2 by site.

All samples were preserved in 70% ethanol, and taken to the laboratory for identification. Finally, animals were sorted, identified, and counted using a dissecting microscope and stored in 70% ethanol.

Results

Morphological and taxonomic remarks on the genus Aoroides

In the family Aoridae in north European waters, the only genus having a male with a gnathopod 1 with a merus prolonged into a very large pointed process is *Aora* Kröyer, 1845 (Chevreux and Fage 1925; Lincoln 1979). Moreover, individuals of the family Aoridae having a large process on the merus of the Gnathopod 1 but without an accessory flagellum on the antennae correspond to the genus *Aoroides*. The mandibular palp is very different in the two genera. In *Aoroides*, palp article 3 is rod-shaped and bears a few terminal and sub-terminal long setae, whereas in *Aora*, the palp is elongate ovoid and the posterior margin bears a comb of short setae in addition to several long pectinate setae (Gouillieux et al. 2015). One species of *Aoroides* lacks a mandible palp. It is also a very distinctive feature that *Aoroides* species are typically around 4.0 mm and are therefore much smaller than *Aora* species, which are usually around 6–9 mm.

Three *Aoroides* species had been recently recorded from the French Atlantic coast of the Bay of Biscay from Brest to the North to Hossegor in the south (Gouillieux et al. 2015). There were also new records from European waters for these species, which originated from Asia and the Northwestern Pacific (Gouillieux et al. 2015). Following the identification key of adult male *Aoroides* from the French Atlantic coast (Gouillieux et al. 2015), two species have been identified in our collection. The antennae 1 of both species have no accessory flagellum. The first species, *A. longimerus*, has gnathopod 1 that is densely setose, and the second species, *A. semicurvatus*, has a gnathopod 1 that is glabrous and outer ramus of uropod 3 marginally bare and inner ramus with 1–2 spines (Figure 2C, D).

Hydrological conditions during the sampling

On 12 February 2019, the sea temperature in the Granville harbour was 7.2 °C, with a salinity of 33.6, a turbidity of 8.1 NTU, and an oxygen concentration of 6.8 mL.L^{-1} . On 25 February 2019 the sea temperature at the Vauban Basin (Le Havre harbour) was 7.2 °C, with a salinity of 26.6, a turbidity of 0.8 NTU, and an oxygen concentration of 7.1 mL.L^{-1} . On 1 March 2019 the sea temperature in the Cherbourg harbour was 9.2 °C, with a salinity of 33.8, a turbidity of 1.2 NTU, and an oxygen concentration of 6.5 mL.L^{-1} .

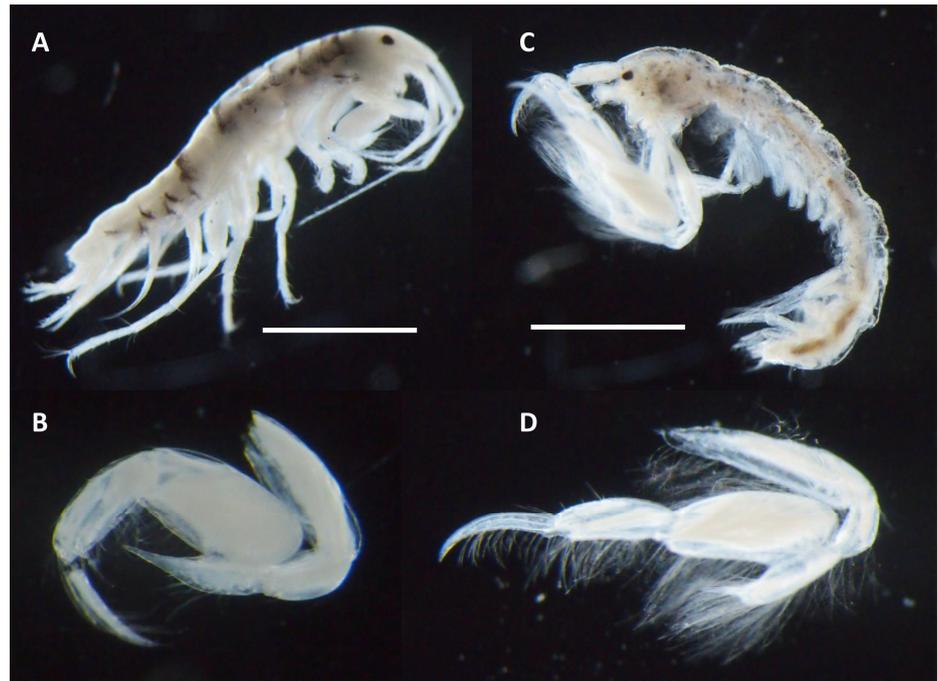


Figure 2. Photographs of *Aoroides semicurvatus* male specimen (MNHN-IU-2019-2544): A) right lateral view, B) right outer gnathopod 1 view; *Aoroides longimerus* male specimen (MNHN-IU-2019-2543): C) left lateral view, D) left outer gnathopod 1 view. Scale bar = A, C: 2 mm; B, D: 0.2 mm. Photo by Jean-Philippe Pezy.

Table 1. Number of *Aoroides* males recorded between February and April 2019 along the Normandy coast, French English Channel.

Locality	Latitude	Longitude	Substratum	Date	Number of <i>A. longimerus</i>	Number of <i>A. semicurvatus</i>
Granville	48.83339	-1.600202	Pontoon	02/12/2019	6	2
Cherbourg	49.647825	-1.621706	Pontoon	03/1/2019	66	–
Saint-Vaast-La-Hougue	49.572982	-1.271393	<i>Sargassum muticum</i>	04/16/2019	4	1
Le Havre (Vauban basin)	49.490042	0.123748	Pontoon	02/25/2019	18	–

New records

As for other Aoridae species, only the males were identified (Table 1). *Aoroides longimanus* males was recorded on pontoons in three marinas of the Normandy zone: Granville, Cherbourg and Le Havre (Vauban Basin), and on one site colonized by the algae *Sargassum muticum*: Saint-Vaast-La-Hougue (Table 1). Males among the 94 individuals varied in length from 3.5 to 5 mm and possessed a gnathopod 1 that was very setose (Figure 2). Four males have been deposited at the Museum National of Natural History at Paris (MNHN-IU-2019-2543).

The three individuals of the second species, *Aoroides semicurvatus*, were recorded in two sites: Granville marina and Saint-Vaast-La-Hougue in *Sargassum muticum* algae. Males varied in length from 3 to 4.5 mm and possessed a gnathopod 1 that was poorly setose (Figure 2A). One male has been deposited at the Museum National of Natural History at Paris (MNHN-IU-2019-2544).

In Saint-Vaast-La Houge, the density of *A. longimerus* was 0.44 individuals per m², while it reached 234 ind.m⁻² at Le Havre, 218 ind.m⁻² at Cherbourg and only 29 ind. m⁻² at Granville. The density of *A. semicurvatus* was lower, from 0.1 ind.m⁻² at Saint-Vaast-La-Hougue to 6 ind.m⁻² at Granville.

Discussion

As assumed by Gouillieux et al. (2015), we expected that both these species *Aoroides longimerus* and *A. semicurvatus* were unintentionally introduced from the Atlantic to Normandy through oyster transfers from French centers of oyster *Crassostrea gigas* productions. Saint-Vaast-la-Hougue is an important hot spot in Normandy oyster production where there is continually on-going transfer between centers of oyster productions between the Bay of Biscay and the English Channel. It is known that bivalve aquaculture and transfer activities were important in dispersal and colonization of new zones for NNS (Brenner et al. 2014). As an illustration, the amphipod NNS *Grandidierella japonica*, was also unintentionally introduced in the Arcachon Bay in 2013 through oyster transfer (Lavesque et al. 2014). This mode of introduction in Normandy was also expected for this species that has been recorded along the Calvados coast in the eastern part of the English Channel since some years (Baffreau et al. 2018).

Species of the family Aoridae were mainly found in the intertidal zone and shallow waters (< 50 m) and often associated with algae and sessile fauna especially hydroids and in oyster beds (Lincoln 1979; Ariyama 2007; Myers 2009). The presence of both species on marina pontoons was probably due to the presence of important biofouling on these floating structures. Barnacles, ascidians, the polychaete *Ficopotamus enigmaticus* (Fauvel, 1923) at Honfleur, and the bivalve *Mytilus edulis* (Linnaeus, 1758) at Le Havre Vauban basin, dominated the sessile fauna, while the amphipods *Monocorophium acherusicum*, *M. sextonae*, and the tanaisids *Heterotanais oerstedii* (Kröyer, 1842) and *Zeuxo holdichi* Bamber, 1990, dominated the vagile fauna (*unpublished data*). Nevertheless, *Aoroides longimerus* and *A. semicurvatus* were present only in three marinas, i.e. Granville, Cherbourg and Le Havre, of the eight sampled during the winter 2019 (Figure 1). In a previous study on biofouling reefs formed by the polychaete species *Ficopotamus enigmaticus* in Normandy marinas, Charles et al. (2018) had not reported the presence of both these species in winter 2015 (January to March), while they were common in three marinas Cherbourg, Ouistreham and Honfleur in the 2015 and 2019 samplings.

We expect that the two species had recently appeared in Normandy and that they were in a colonisation phase of most of the favourable habitats for these species, mainly the marina pontoons. After a phase of colonisation by aquaculture, transfer by biofouling could be evoked as another pathway explaining the presence of both species of *Aoroides* in the English Channel

harbours such as Granville, Cherbourg and Le Havre. These three harbours are characterized by international traffic of merchant vessels mainly between France and United Kingdom for Granville and Cherbourg and worldwide for Le Havre. The Le Havre harbour was a major source of introduction of NNS into the English Channel, with a large number already detected (Breton 2014; Pezy et al. 2017, 2020). Furthermore, both of the harbours at Le Havre and Cherbourg are also predisposed to biological invasions since they exhibit several major anthropogenic activities that can be responsible for NNS introductions such as those relating to recreational and commercial harbours (Baffreau et al. 2018). Moreover, Zabin et al. (2014) had noted that some recreational boats can be used to travel long distances, and their relatively low speeds make them ideal vectors for those species that attach to their hulls, enabling them to contribute to NNS dispersal via marinas and harbours. Similarly, Ulman et al. (2017), based on sampling of 34 Mediterranean marinas during a one-year survey in 2015–2016 from Spain, France, Italy, Malta, Greece, Turkey and Cyprus, had shown that marinas acted as major hubs for the transfer of marine NNS, and that recreational boats acted as effective vectors of spread.

For the moment, both processes of spread of these two *Aoroides* species in Normandy could be suggested: oyster transfer between production centers of *Crassostrea gigas* along the Atlantic and French English Channel coast, followed by biofouling on recreational and commercial vessels. In future, DNA barcoding studies must be used to identify the origin of these species in the Normandy waters and to determine primary and secondary introductions. For example, this molecular method was used to identify the origin of *Paracaprella pusilla* Mayer, 1890, in European and Mediterranean waters (Cabezas et al. 2019) and other amphipods in European waters, e.g., *Caprella mutica*, *Caprella scaura*, *Ampithoe valida* Smith, 1873, and *Jassa marmorata* Holmes, 1905 (Ashton et al. 2008; Cabezas et al. 2014; Pilgrim and Darling 2010). As underlined by Marchini and Cardeccia (2017), amphipod NNS offer a large spectrum of future taxonomic research into cryptogenic species, unresolved taxonomy and overlooked introductions. The elucidation of the origin and dispersion of *Aoroides* species in European waters will be studied in the future, similar to that of the caprellid *Paracaprella pusilla* in European and Mediterranean waters (Cabezas et al. 2019), which represented a collaboration between observers, naturalists and geneticists of marine species.

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