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**Rapid Communication****The American protobranch bivalve *Yoldia limatula* (Say, 1831) in European waters**

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**OPEN ACCESS****Abstract**

Three specimens of the Northern American bivalve *Yoldia limatula* (Say, 1831) were retrieved from Le Havre Harbour (The English Channel, Normandy, France) for the first time in 2021. This species was initially recorded from Europe in the Netherlands in 2019, after which specimens were found in Belgium in 2020. This is the first record of this species in Normandy and France now extends the invasive range of the species from the eastern part of the English Channel to the southern part of the North Sea. This species is considered as non-indigenous in Normandy and was most likely introduced via ballast water due to intense marine shipping between North American and European harbours.

**Key words:** bivalve, Non-Indigenous Species, Le Havre harbour, English Channel

**Introduction**

The census of introduced marine species is currently a major objective in several marine regions, whether in the Mediterranean or in the Baltic where lists of non-indigenous species (NIS) are regularly published (see Zenetos et al. 2012). Descriptor 2 of the Marine Strategy Framework Directive (MSFD) is dedicated to NIS, i.e. species introduced through human activities not yet not disturbing ecosystem functioning (Boon et al. 2020; Tsiamis et al. 2020). Species introduced voluntarily or involuntarily outside their native distribution range may not exhibit an invasive character. Noticeably, only a small number of species may find favourable ecological conditions to settle and proliferate in the colonized area without predators to regulate their populations (Stiger-Pouvreau and Thouzeau 2015).

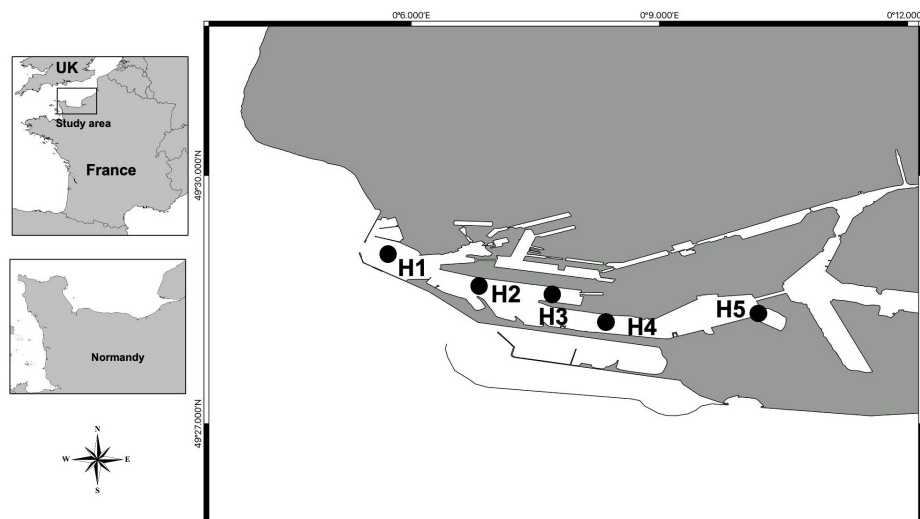
The record of new NIS is most often fortuitous even if there are studies dedicated to their survey in areas favourable to their introduction and settlement such as harbours and marinas, where many species were introduced by ship ballast water (Gouletquer et al. 2002; Minchin et al.

2013; Stiger-Pouvreau and Thouzeau 2015; Gouletquer 2016; Pezy et al. 2021). Professional or recreational fishermen often catch or harvest species unknown to them; providing unique opportunities to record the presence of new NIS (Pezy et al. 2019a, b). Furthermore, scientific or biomonitoring programs aiming at assessing the effects of human activities on benthic habitats may also lead to the sampling of new NIS (Pezy et al. 2021).

During a sampling survey part of the scientific project “Foram-INDIC” intended to compare foraminiferal and macrofaunal benthic communities in transitional waters in the eastern part of the English Channel, three specimens of the bivalve species *Yoldia limatula* (Say, 1831), yet unrecorded in France, were found in Le Havre Harbour basins. It is commonly called the file yoldia, from the family Yoldiidae, common along the Atlantic coast of North America, from the Gulf of St. Lawrence to New Jersey. Lists of NIS species established by Breton (2014) and Pezy et al. (2021) showed that the Havre harbour is one of the main spot of introduced species in Normandy, France and North-European waters. Specifically, several new NIS were identified during the last decade in the Havre basin and in the neighbouring Seine Estuary: the established populations of the polychaete *Euchone limnicola* Reish, 1959 (Guyonnet 2015), the sponge *Celtodoryx ciocalyptoides* (Burton, 1935) (Berno et al. 2016), the mysid *Neomysis americana* (S.I. Smith, 1873) (Massé et al. 2018; Pezy et al. 2019c), the amphipod *Aoroides longimerus* Ren & Zheng, 1996 (Dauvin et al. 2020), the isopods *Paranthura japonica* Richardson, 1909 (Pezy et al. 2020) and *Ianiropsis serricaudis* Gurjanova, 1936 (Raoux et al. 2020), and the unique records of the prawns *Penaeus japonicus* Spence Bate, 1888, *Penaeus semisulcatus* (De Haan, 1844 in De Haan, 1833–1850) (Pezy et al. 2017a), and the tropical crab *Guinearma alberti* (Rathbun, 1921) (Pezy et al. 2017b).

One specimen of *Yoldia limatula* was previously recorded in European waters in the Western Scheldt (Netherlands) on September 26 2019 at a depth of 18 m (Driessen et al. 2020; <https://www.naturetoday.com/intl/nl/naturereports/message/?msg=26297> accessed July 15<sup>th</sup> 2021). One year later de Kisangani and Kerckhof (2020) reported the finding of three specimens of *Y. limatula* on the Belgian beach of Koksijde, after a storm. Originating from North-America, this is the first reported NIS case of a protobranch bivalve; compared to ca. 200 species in other bivalve subclasses (WORMS consulted on 1 October 2021). Species from this class are the most primitive bivalves, lacking the lamellibranch gill that enables filter-feeding in most other bivalves. Most species of the subclass are found in deep-sea waters.

The aim of this record is 1) to describe the three sampled specimens recorded in the Havre harbour together with the ecological features of the benthic habitats where they were found and 2) to discuss the hypothetical introduction pathway of this new NIS in the French waters.



**Figure 1.** Map of the Le Havre basin with the location of the five samplings stations.

**Table 1.** Main characteristics of environmental parameters at both stations H3 and H4 from the Le Havre harbour basins.

	Station H3	Station H4
GPS coordinates Latitude	49.476307N	49.469837N
GPS coordinates Longitude	0.127322E	0.139166E
% of clay (< 2 $\mu\text{m}$ )	0.40	0.70
% of silt (2–63 $\mu\text{m}$ )	96.20	95.60
% of sand (>63 $\mu\text{m}$ )	3.40	3.70
C <sub>org</sub>	1.11 $\pm$ 0.01%	1.17 $\pm$ 0.02%
N <sub>org</sub>	0.03 %	0.03 %
C/N ratio	37.3 $\pm$ 0.9	37.4 $\pm$ 0.4

## Materials and methods

### Study area

Three specimens of *Yoldia limatula* identified within the *Yoldia* species diagnosis of Ockelmann (1954) and Abbott (1974) were found on the 20<sup>th</sup> of April 2021 in two stations out of the ten sampling stations in Le Havre harbour: H3 and H4. Stations H3 and H4 are located in the upper part of the commercial basins of the Le Havre harbour (Figure 1; Table 1). The samples of Le Havre harbour (H) were collected on the 11<sup>th</sup> of September 2019 and on the 20<sup>th</sup> of April 2021. The stations were along the dykes in areas without recent dredging; at water-depth between 4–8 m at low tide along a gradient from the entrance of the Harbour (station H1) to the station H5 in the inner part of the basin.

### Sampling strategy

At each sampling station, sediment and macro-invertebrates were sampled using a 0.1 m<sup>2</sup> Van Veen grab. Four replicates were taken, three for the fauna and one for sediment parameters. After sieving the sediment on a 1-mm mesh, all organisms collected were fixed with neutral formalin buffered at 10% in the laboratory before being sorted.

### *Grain-size analysis*

To assess sediment granulometry, laser diffraction particle-size analysis was carried out (parameters used: measured interval: 0.02  $\mu\text{m}$  to 2 mm, real part: 1.54 and imaginary part: 0.1, mie diffraction method).

### *Organic matter analysis*

Samples for organic matter (OM) analysis were dried for three days at 60 °C and then burned at 500 °C for 4 h. Sediment samples for organic carbon analyses were preserved at -20 °C at the laboratory. Total organic carbon and nitrogen contents were obtained after sediment grinding by hand, by quantifying the total amount of carbon and nitrogen (measured in dried samples). The amount of inorganic carbon and nitrogen (measured in samples burned at 550 °C for 5 hours) was subtracted. It was determined with an elemental analyzer (ThermoFisher Flash 2000, Laboratory of Oceanology and Geosciences in Wimereux-France) and expressed as the % of  $C_{\text{org}}$  and  $N_{\text{org}}$  per total weight of dry sediment. The C/N ratio was calculated at each station to determine the terrestrial or marine origin of the organic matter.

## **Results**

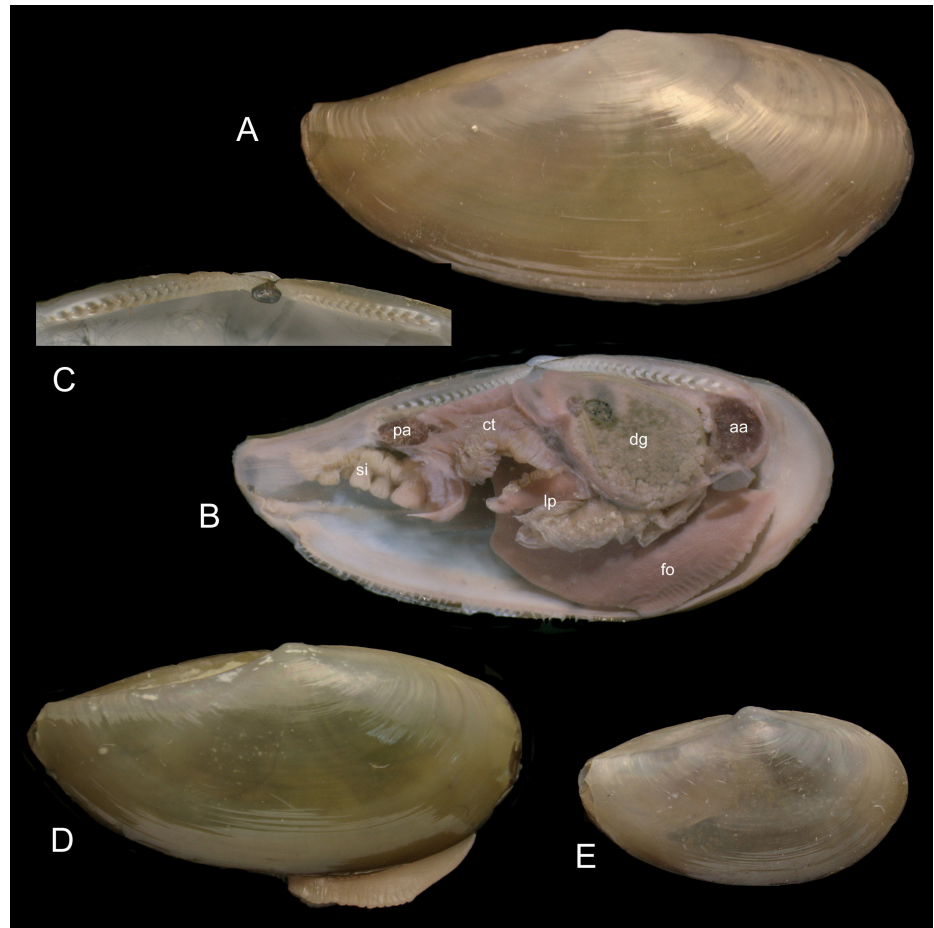
### *Yoldia limatula record in Le Havre harbour basins*

*Yoldia limatula* was absent from collections in 2019. In April 2021, three individuals were sampled from stations H3 (one specimen) and H4 (two specimens) in the inner part of the harbour (Figure 1).

In April 2021, seawater environmental parameters at both stations were exactly the same: salinity: 32, sea temperature 9.9 °C, Chlorophyll *a* 0.65  $\mu\text{g/L}$  and turbidity 7.75 NTU. Sediment parameters, including grain size, at both stations were similar (Table 1). Organic matter (by loss-on-ignition) content in the sediment was 1.15 % (Table 1).

### *Main characteristics of the three recorded individuals*

*Yoldia limatula* cannot be confused with any member of the native fauna of the temperate Eastern Atlantic (see discussion for more detailed comparisons with similar species). The maximum shell length of the specimens collected here was 25.1 mm (Figure 2), but individuals can reach over twice that size in its native area. The shell shape was elongate, tapered posteriorly and rounded anteriorly, and very compressed (i.e., the width is about half of the dorso-ventral height). The umbo was near the vertical midline. The outer surface was smooth, except for marked growth lines, and covered by an extremely glossy, greenish brown periostracum. The interior of the valves was whitish, not nacreous. The hinge was taxodont, interrupted by a conspicuous internal ligament situated beneath the umbo,



**Figure 2.** *Yoldia limatula* (Say, 1831) from Le Havre harbour, Théophile Ducrocq basin, 4–8 m depth. A. Specimen 25.1 mm long, viewed from right side. B. Same, with right valve and right mantle lobe removed (aa: anterior adductor muscle; ct: right ctenidium; dg: digestive gland; fo: foot; lp: right labial palp; pa: posterior adductor muscle; si: siphon). C. Same, hinge of the right valve showing the internal ligament and taxodont teeth. D. Specimen 18.5 mm long, with protruding foot. E. Specimen 7.5 mm long. Photograph by Serge Gofas.

with series of chevron-shaped teeth anteriorly (ca. 18 teeth) and posteriorly (ca. 15 teeth) to the ligament. The animal had a large foot, striated laterally, and large labial palps. The ctenidium, situated posterior to the visceral mass, was small and pectinate. The anterior adductor muscle was about twice as large as the posterior one.

#### *Associated macrofauna*

In April 2021, the dominant species at both stations were the polychaetes *Euchone limnicola* Reish, 1959, *Heterocirrus* spp. and *Melinna palmata* Grube, 1870, and the bivalves *Abra alba* (W. Wood, 1802), *Corbula gibba* (Olivi, 1792), and *Nucula nitidosa* Winckworth, 1930. This community corresponds to the EUNIS classification MB6244 *Melinna palmata* with *Magelona* spp. and *Thyasira* spp. from Atlantic infralittoral sandy mud habitats (see <https://www.eea.europa.eu/data-and-maps/data/eunis-habitat-classification/habitats>).



## Discussion

The genus *Yoldia* comprises several shallow-water, continental shelf species mostly distributed in high latitudes of the Northern Hemisphere (Ockelmann 1954; Abbott 1974). The species found in this study, *Yoldia limatula*, has a cold-temperate native range from the Gulf of Saint Lawrence to North Carolina in the Western Atlantic. In comparison, the similar species *Yoldia hyperborea* (Gould, 1841), is common in European Arctic seas (Dautzenberg and Fischer 1912) and has its type locality in Massachusetts, close to its southern limit. An early report of *Y. limatula* from Northern Norway (Sars 1878) was later identified as *Y. norvegica* Dautzenberg and Fischer, 1912, which was eventually classified as a subspecies of *Y. hyperborea* by Ockelmann (1954). *Yoldia hyperborea* differs from *Y. limatula* in having a blunt, not tapering posterior end and a sinuous, not evenly rounded antero-ventral edge. The other similar boreal species from the Western Atlantic are *Y. sapotilla* (Gould, 1841) and *Y. myalis* (Couthouy, 1839). Both of these species are shorter than *Y. limatula* (i.e., the shell length is less than twice the dorso-ventral height); in addition, the umbo of *Y. myalis* is closer to the posterior end, not median. Based on morphological differences, the three bivalves sampled in Le Havre harbour clearly belong to the species *Y. limatula*. This paper, hence, reports the first record of the North-American bivalve *Y. limatula* in France, specifically in Le Havre harbour basins. This harbour sits at the entrance of the Seine estuary in the eastern part of the English Channel. We hypothesize, due to the distance between this location and its native range, that these organisms have been introduced to Le Havre harbour.

Protobranch bivalves have relatively small, short-lived, non-feeding larvae known as “pericalymma” that have a ciliated epithelial test in which the definitive adult structures develop. The pericalymma has been described for *Yoldia limatula* (Drew 1898, 1899a, b) but also for other protobranch bivalves (see Gustafson and Reid 1986 and references therein). The eggs of *Y. limatula* are ca. 150 µm in size, and the test is composed of 42 cells organized in five rows. The middle three rows are ciliated, which enables swimming. After 90 to 120 hours (4–7.5 days), the test is cast apart, and the larva settles on the bottom (Drew 1898, 1899a, b). This planktonic stage allows limited dispersal under natural conditions but is compatible with a transatlantic route of a commercial ship (a route from New York to Le Havre is covered in seven days, see <https://www.freightercruises.com/18/03/Liberty.pdf>).

In Le Havre harbour, specimens were found at shallow water depth (< 20 m) in muddy soft-bottom sediment near estuarine environments, similarly to what is observed in its native range. Noticeably, in Casco Bay, Maine, their principal habitat is reported as “in the shallow coves and inlets, where soft mud has accumulated. They are most abundant in water

from one to five fathoms depth, and probably never occur above low tide mark” (Drew 1898). This habitat is favoured because the species is a subsurface and surface deposit-feeder. Its feeding behavior enhances sediment resuspension through expulsion of loose pseudofaeces directly into the water column (Blender and Davis 1984).

Size frequency analysis of *Y. limatula* from the Bideford River, Malpeque Bay, Prince Edward Island, Canada, during the summers of 1979 and 1980 showed that the size range 25 to 35 mm shell length was dominant from May to July (Lewis et al. 1982). Recruitment occurred in August, as 50% of the population was < 5 mm (Lewis et al. 1982). Shell length in this species can be correlated with age. Bivalves between 13.8 and 17.7 mm are one-year-old, between 26.7 and 28.0 mm are two years old, and between 33.8 and 35.2 mm are three years old; individuals larger than 35.2 are four year old and have a mean length of 38.7 mm (Lewis et al. 1982). In Le Havre harbour, identified specimens measured 7.5, 18.5 and 25.1 mm, and are classified as one-year old specimens according to the length given by Lewis et al. (1982). Therefore, the introduction of this species in the English Channel and French coastal water is hypothesized to have occurred recently as only young bivalves were present in April 2021 but absent in the same station in September 2019.

This is the third record of *Y. limatula* in the European waters, outside its native range, but all records have been observed in only the last three years (since 2019). Introduction in the southern part of the North Sea probably occurred *via* ship ballast waters from the eastern coast of North-America, where the species has been recorded from Nova Scotia to North Carolina. The single specimen reported from the Western Scheldt, Netherlands on muddy sediment at 17.7 m depth in September 2019 was 48.8 mm in length (Driessen et al. 2020). The three Belgian specimens (46, 47- and 48-mm length) were discovered after a storm on the beach of Koksijde at the end of September 2020 and the beginning of October 2020 (De Kisangani and Kerckhof 2020). All of the previous North Sea specimens were adults, probably more than four years old, according to Lewis et al. (1982). Therefore, the Belgium-Netherlands populations could have been introduced as far back as the mid-2010s and could be the origin of the Le Havre population surveyed here *via* ballast water transport, suggesting secondary spread. Although speculative, it may also be hypothesized that the species arrived directly from North-American waters to the English Channel and the south of the North Sea *via* multiple introductions. Of the 75 non-indigenous species (NIS) recorded in the Le Havre harbour basins, two-thirds are suspected of being introduced *via* ballast water (Pezy et al. 2021). Le Havre harbour remains a hot spot for the introduction of NIS in Normandy and the English Channel, and about 50% of the NIS recorded in Normandy by Pezy et al. (2021) were recorded in this location.



The present study was the third record in three successive years of *Yoldia limatula* in European waters. At this stage, the record of only three young specimens of *Y. limatula* does not allow us to confirm its NIS status along the coast of Normandy. Larger surveys are necessary to further describe its distribution in European waters and determine the existence of established populations outside its native range in the North-America Atlantic, particularly in Le Havre harbour.

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## Authors' contribution

Research conceptualization: V.M.P. Bouchet, J.C. Dauvin and J.P. Pezy. Sampling campaign: V.M.P. Bouchet, J.C. Pavard and J.P. Pezy. Species identification: J.C. Dauvin and S. Gofas. The Figure 1 was produced by J.P. Pezy while S. Gofas executed the Figure 2. Editing of the typescript: all co-authors.

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