

## Research Article

## First record of the invasive brackish water mytilid *Limnoperna securis* (Lamarck, 1819) in the Bay of Biscay

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Received: 2 March 2011 / Accepted: 17 June 2011 / Published online: 30 June 2011

### Abstract

The occurrence of the non-indigenous species *Limnoperna securis* belonging to Mytilidae family is recorded for the first time in the Bay of Biscay. Numerous individuals were collected in intertidal and shallow waters in the inner part of Nervión estuary (Bizkaia, Basque Country, SE Bay of Biscay). In the present paper, notes about the history of this small brown mussel invasion, vectors of introduction and dispersal, as well as consequences of invasion are discussed.

**Key words:** invasive species, biological invasion, vectors, ecological traits, Nervión estuary, Mytilidae

### Introduction

Maritime traffic across the oceans and culturing of non-native organisms at the edge of the sea contributed to the spread and establishment of an ever increasing number of alien species in coastal and brackish water environments (Reise et al. 1999).

This phenomenon, a major threat to biodiversity of ecosystems, is not strange to the Basque coast. The great increase of naval transport and marine economic activities in the Bay of Biscay in the last decades has been responsible for the displacement of many exotic species. Since the 1970s an important number of alien species, specially from Mediterranean, tropical and sub-tropical areas have been recorded in the Basque coast (Ibáñez 1980; Ibáñez and Salo 1975; Ibáñez and Motos 1977; Ibáñez et al. 1986; Casares 1977; Casares et al. 1987; Gorostiaga and Limia 1985; Gorostiaga et al. 1981, 1988; Ceberio et al. 1998; Secilla et al. 1997, 2007; Santolaria et al. 1998; Martínez and Adarraga 2005a, b, 2006a; Martínez et al. 2002, 2006b).

The two last species detected belong to the Mytilidae family, the Asian mussel *Musculista senhousia* (Benson in Cantor, 1842) and the small brown mussel *Limnoperna securis*

(Lamarck, 1819). The first one was reported by Bachelet et al. (2008) in the Bidasoa estuary, whereas the second one is presented here.

*Limnoperna* is one of more than 30 genera in the Mytilidae family, which includes a group of relatively small bivalve molluscans. To date, the *Limnoperna* genus comprises nine valid species: *L. atrata* (Lischke, 1871), *L. balani* (Ockelmann, 1983), *L. fortunei* (Dunker, 1857), *L. hepatica* (Gould, 1850), *L. inconstans* (Dunker, 1856), *L. mangle* (Ockelmann, 1983), *L. pulex* (Lamarck, 1819), *L. sambasensis* (Dautzenberg, 1903) and *L. securis*.

In recent years, both *L. fortunei* and *L. securis* have significantly spread, increasing their geographical distribution. Specifically, *L. securis* has established populations in the Indian Ocean: Kimura et al. (1999), Kohama et al. (2001), Shirafuji and Sato (2003); Mediterranean: Lazzari and Rinaldi (1994), Sabelli and Speranza (1994) as *Xenostrobus* sp., De Min and Vio (1997), Russo (2001), Giusti et al. (2008), Zenetos et al. (2010), Barbieri et al. (2011); and in the Atlantic Ocean: Garci et al. (2006), Pascual et al. (2010) (Table 1).

In its natural habitat this suspension filter feeder is a common species in eutrophic estuaries. It is present in great quantities on timber, stones or dead shells in upper reaches of

estuaries where the water may be almost fresh for a considerable part of the year. The species is not found in the sea or in the downstream parts of estuaries where the salinities are constantly high (Wilson 1967).

The Nervión estuary, situated in the province of Bizkaia, is the metropolitan main axis of Bilbao, and therefore, has been subjected to a large pollutant load from domestic and industrial sources, causing gross impairments to its biological system (González-Oreja and Saiz-Salinas 1999).

In 1979 the Consorcio de Aguas de Bilbao-Bizkaia (the local authority responsible for water supply and wastewater treatment) launched a plan to overcome this situation, approving a sewerage scheme for the treatment of waste and polluted land. To assess the results of these measures on the environmental quality of the estuary, an annual monitoring programme (including physico-chemical and biological parameters in the waters and sediments) was established in 1989 by the Consorcio and carried out by AZTI (Borja et al. 2006).

In a 2010 survey, some individuals of *L. securis* were identified in the innermost part of the estuary. This record extends its geographical distribution and enlarges the list of alien species that have arrived into the Bay of Biscay.

## Materials and methods

### Study area

The Nervión River, which runs through the city of Bilbao, is located in the northeast of the Iberian Peninsula flowing into the Cantabrian Sea (Bay of Biscay). The final tract, the zone of marine influence, is known as the Nervión estuary. This estuary is 22 km long and communicates with the sea through the Port of Bilbao.

The natural features of the estuary have been modified dramatically by urban, industrial and port settlements, which have occupied practically the whole of the valley during the last 150 years (Cearreta et al. 2004). As a result, the original estuary has been reduced in size through land reclamation to form a tidal navigation channel from the city of Bilbao to the open sea (average dimensions: 100 m wide, <10 m deep) and this was completed in 1885 (Junta de Obras del Puerto 1910).

Nowadays, the Nervión estuary has a surface of 24 km<sup>2</sup> with an approximate volume of 200 10<sup>6</sup> m<sup>3</sup>. In the outer part it has an average depth of 25 m, with a maximum of 30 m; ranging between 5 and 10 m in the inner part.

The areas where *Limnoperna securis* was found are located in the inner part of the estuary (Figure 1).

### Methodology

In the year 2010, a total of 13 sampling stations between 1 and 30 m deep were selected along the longitudinal axis of the Nervión estuary, from the inner to the outer reaches. Later, in 2011 another two sampling stations were selected in the intertidal area.

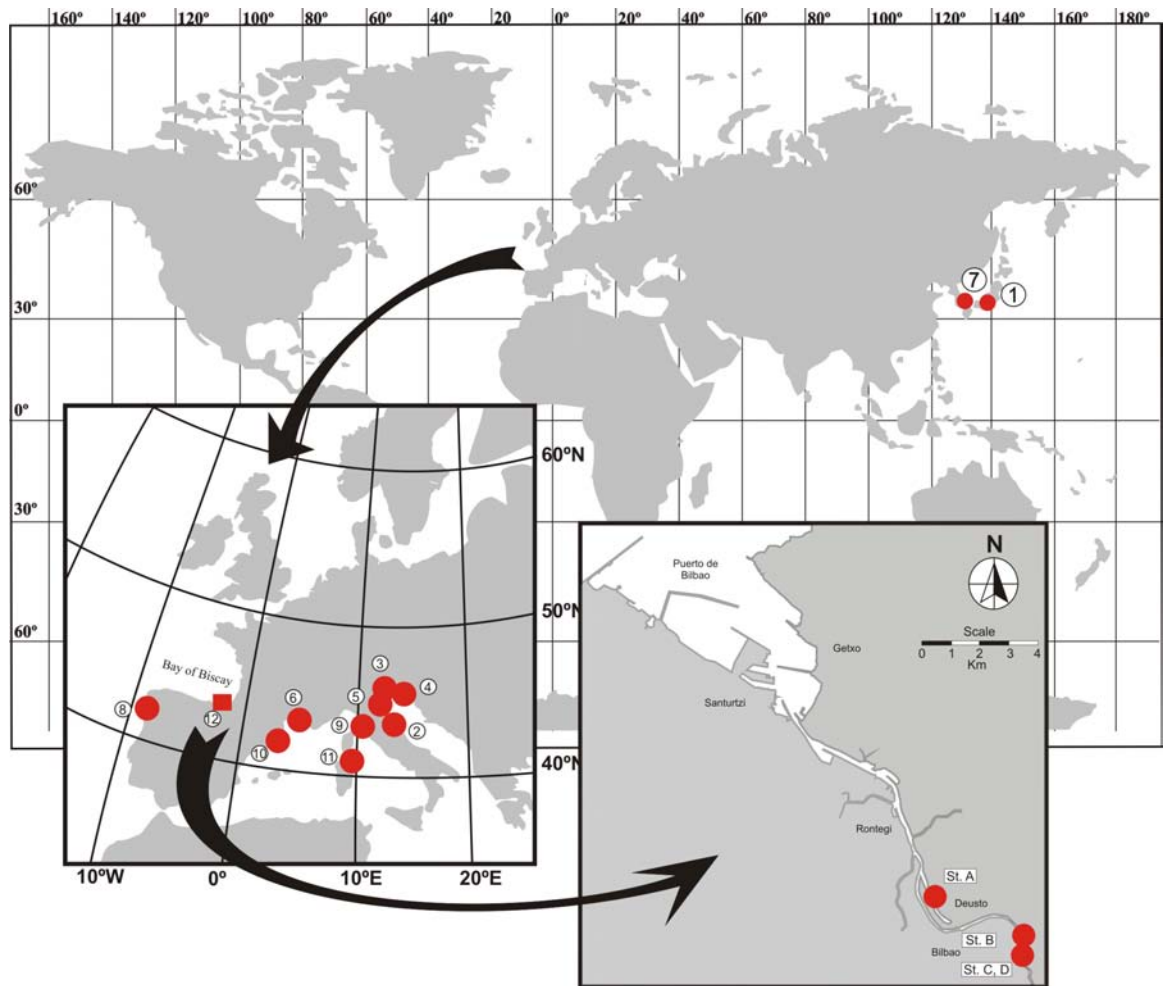
Subtidal samples were taken on 6 October 2010 by AZTI. At each station, three replicates were collected using a Van Veen dredge (0.1 m<sup>2</sup>); two for the macrobenthic communities characterization and one for sediment analysis. In the two benthic replicates, the sediments were washed through a 1 mm mesh sieve and fixed with a 4% formalin solution stained with rose Bengal. In the laboratory, the macrofauna were sorted and transferred to 70% ethanol.

Intertidal samples were taken on 20 February 2011 by the authors. An area was selected as a sampling station where individual density seemed to be the highest in the estuary (Figure 2a). Samples were taken at two tidal sites, at 1.0 m (Station C) and 1.5 m (Station D) above sea level (Figure 2b). Station C was sampled on a reef of serpulids *Ficopomatus enigmaticus* (Fauvel, 1923); whereas rocky substrate was chosen for the Station D. Samples were collected scraping a surface of 400 cm<sup>2</sup> (20 × 20 cm). Immediately after collection, samples were washed through a 1 mm mesh sieve, fixed in a 5% formalin solution, preserved in 70% ethanol, and subsequently processed in the laboratory, including determination of the species community composition.

The identification and description of the *L. securis* specimens was based on the morphological descriptions provided by Wilson (1967).

Observations and measurements were made using an Olympus stereomicroscope and a calibre. Individuals were photographed on an Olympus stereomicroscope with a Nikon D50 digital camera.

First record of *Limnoperna securis* in the Bay of Biscay



**Figure 1.** Records of *Limnoperna securis* and map showing the study area and the locations of the four stations where this species has been collected.

**Table 1.** Records of *Limnoperna securis* in the world.

Map ref.	Location/Country	Record coordinates		First observation	Reference
		Latitude	Longitude		
1	Kojima Bay, Japan	34°38'N	134°02'E	1972	Kimura et al. 1999
2	Ravenna lagoon, Italy	44°27'N	12°16'E	1992	Lazzari and Rinaldi 1994
3	Venice lagoon, Italy	45°25'N	12°20'E	1992	Sabelli and Speranza 1994
4	Porto Buso, Italy	45°43'N	13°15'E	?	De Min and Vio 1997
5	Delta of Po river, Italy	44°57'N	12°30'E	2001	Russo 2001
6	Etang du Vidourle, France	43°27'N	04°30'E	?	Zenetos et al. 2003
7	Sacheon and Masan, South Korea	35°01'N 35°12'N	128°02'E 128°35'E	?	Shirafuji and Sato 2003
8	Ría de Vigo, Spain	42°20'N	08°36'W	1995	Garci et al. 2006
9	Leghorn harbour, Italy	44°05'N	09°57'E	2006	Giusti et al. 2008
	Scolmatore Canal, Italy	43°35'N	10°18'E	2008	Barbieri et al. 2011
	Navicelli Canal, Italy	43°38'N	10°21'E	2008	Barbieri et al. 2011
	Morto river, Italy	43°44'N	10°17'E	2009	Barbieri et al. 2011
	Arno river, Italy	43°41'N	10°20'E	2010	Barbieri et al. 2011
10	Fluviá river, Spain	42°12'N	03°06'E	2007	Barbieri et al. 2011
11	Gulf of Olbia, Sardinia, Italy	40°55'N	09°30'E	2010	Barbieri et al. 2011
12	Nervión estuary, Spain	43°15'N	02°56'W	2010	This study

## Results

More than 4643 specimens of *Limnoperna securis* were identified in the Nervión estuary. The specimens were collected from two of the thirteen subtidal sampling stations (Stations A and B) and two intertidal stations (Stations C and D). A total of 6228 organisms were identified from these four samples, belonging to 28 species or higher taxa. Mollusca were the most abundant taxonomic group, followed by Annelida and Crustacea (Appendix 1). This is the first record of *L. securis* in the Bay of Biscay (Figure 1).

### Systematics

#### *Limnoperna securis* (Lamarck, 1819)

The nomenclature of this species has been very confusing. *Limnoperna securis* has been reported under a variety of synonyms, as *Modiola securis* Lamarck, 1819, *Modiola vexillum* Reeve, 1857, *Perna confusa* Angas, 1871, *Modiola fluviatilis* Hutton, 1878, *Modiolus pulex* (non Lamarck) Thiele, 1930, *Xenostrobus securis* (Lamarck, 1819) and *Limnoperna fortunei kikuchii* Habe, 1981 (Wilson 1967)

### Native range

New Zealand and Australia

### Material examined

3 individuals, Deusto Canal, station A (43°16' 33.60"N, 02°57'50.09"W), mud sediment, 7.2 m depth, 06/10/2010; 409 individuals, El Arenal, station B (43°15'47.50"N, 02°55'25.05"W), mud sediment, 1.4 m depth, 06/10/2010; 2653 individuals, Abando, station C (43°15'31.20"N, 02°55'36.11"W), rocky substrate, intertidal, 1 m above sea level, 20/02/2011; 1578 individuals, Abando, station D (43°15'31.20"N, 02°55' 36.11"W), rocky substrate, intertidal, 1.5 m above sea level, 20/02/2011.

### Diagnosis

Shell equivalve with periostracum smooth and shining. Outline sub-arcuate (strongly arcuate in the older specimens). Dorsal ligamental margin usually straight, ventral margin variably arcuate, and posterior end rounded. Umbones almost terminal. Shell brown (Figure 2 c); although in older specimens can be dark brown and brown with pale yellow zigzag lines in the younger ones

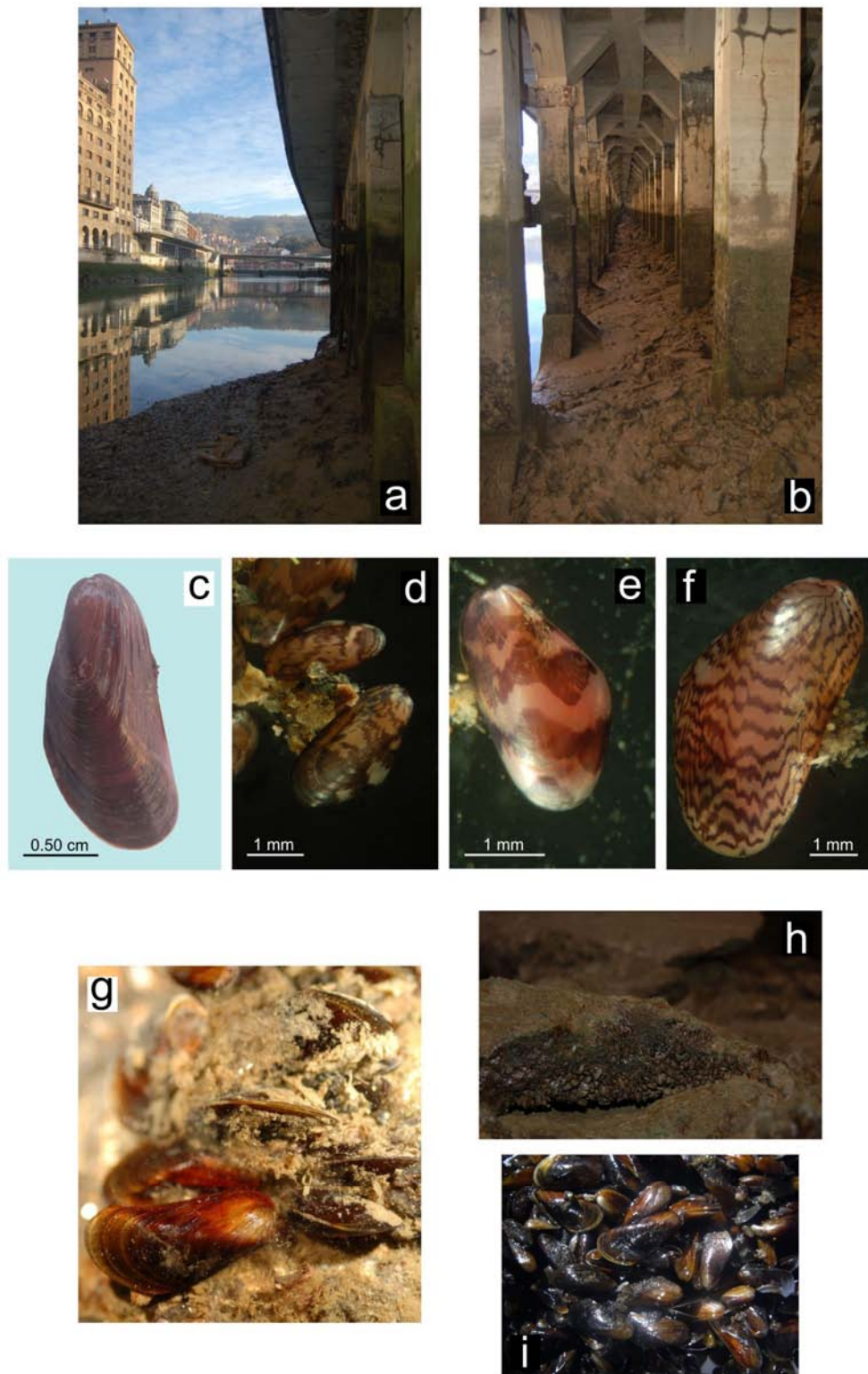
(Figure 2 d, e, f). Interior shell normally purple above and white below the umbonal keel. Size of adults between 20–30 mm. Maximum length found 47 mm.

### Distribution in the Nervión and ecology

In the Nervión, *Limnoperna securis* is restricted to the upper reaches of the estuary. In these areas, it can colonize all kind of substrates; natural as well as artificial. Bathymetrically, it was collected in four stations from the intertidal zone to 10 m depth. Due to the varying environmental conditions in these areas, the benthic communities have been different (Appendix 1).

In shallow waters, *L. securis* was found unevenly, on muddy bottoms. At station A, only 3 individuals (equivalent to 30 ind./m<sup>2</sup>) were obtained at 7.2 m depth; whereas 409 individuals (4090 ind./m<sup>2</sup>) were identified at station B, most of them attached to leaves deposited or buried in the mud at 1.4 m depth. The accompanying macrofauna is characterized in order of abundance by Nematoda; the polychaete *Chaetozone gibber* Woodham and Chambers, 1994; and the bivalve *Corbbula gibba* (Olivi, 1792) in the first case; and by the polychaetes: *Polydora ligni* (Webster, 1879), *Pseudopolydora paucibranchiata* (Okuda, 1937) and *Capitella capitata* (Fabricius, 1780) at the second site.

In the intertidal zone, numerous populations of this small brown mussel have been observed in high concentrations on a great variety of substrates; both natural (rocks, colonies of the tubeworm *Ficopomatus enigmaticus*, branches, trunks and leaves) as well as artificial (walls, tubes, sackcloth, plastic materials, metallic structures, etc) (Figure 2 g, h, i). The densities of *L. securis* were very high here with 2653 individuals counted, within the *F. enigmaticus* reef at 1.0 m above sea level (Station C). As only a 0,20 × 0,20 cm quadrat was sampled, these values imply a density of 66325 ind./m<sup>2</sup>. Lower values, but also abundant, were obtained at Station D, in a dense colony of the same *L. securis* attached to a rock at 1.5 m above sea level. In this case 1578 individuals (equivalent to 39450 ind./m<sup>2</sup>) were collected. The most abundant species at both stations were: *L. securis*, *F. enigmaticus*, Oligochaeta and the isopod *Lekanasphaera rugicauda* (Leach, 1814); and *L. securis*, *F. enigmaticus*, Oligochaeta and the gastropod *Potamopyrgus antipodarum* (Gray, 1843), respectively (Appendix 1).



**Figure 2.** *Limnoperna securis* (from Nervión estuary, Spain) (a) inner part of Nervión estuary; (b) site of intertidal sampling stations; (c) adult; (d, e, f) juveniles; (g) patches of *L. securis* in rocky; (h) colony attached to a stone; (i) specimens of *L. securis* collected at Station D (Photographed by Julián Martínez).

## Discussion

The presence of *Limnoperna securis* in the Basque coast is unclear. *L. securis* is the only brackish water species of this genus that has broadened its geographic range. The first record outside its natural range was in the Kojima Bay (Japan) in 1972 (as *Limnoperna fortunei kikuchii*) (Kimura et al. 1999). Since then, and chronologically, it has been recorded in the Adriatic Sea: Ravenna lagoon (Lazzari and Rinaldi 1994), Venice lagoon as *Xenostrobus* sp. (Sabelli and Speranza 1994), Porto Buso (De Min and Vio 1997), delta of the Po river (Russo 2001); French Mediterranean lagoons, in the Etang du Vidourle area (Zenetos et al. 2003); Sacheon and Masan in South Korea (Shirafuji and Sato 2003); Atlantic waters, in the Ria de Vigo (Galicia) (Garci et al. 2006); and western Mediterranean, in northern Catalonia (Barbieri et al. 2011) and Central Tyrrhenian Sea (Giusti et al. 2008, Barbieri et al. 2011).

Although the potential vectors of spread include: surface currents, aquaculture, ballast waters or ship hulls, the characteristics and distances between these localities suggest that aquaculture and ships transport are the principal vectors for the introduction of *L. securis*.

According to Kimura et al. (1999), this mytilid was first introduced to the Japanese coasts probably from Australia or New Zealand in the 1970s. The introduction was accidental, most likely via shipping, either with ballast water or as hull fouling on ships.

Moreover, its introduction into the Mediterranean and Galicia appears to be related to aquaculture (Occhipinti-Ambrogi 2000; Garci et al. 2006). The great development of mollusc aquaculture (oysters, mussels and clams, traditionally) in these areas seems a likely trans-oceanic vector of invasions of *L. securis* from the Indo-Pacific region. All records refer to locations close to shellfish farming areas, mainly oysters. The transfer of oysters between distant coasts is considered to be one of the major gateways of marine introductions (Reise et al. 1999). After the first arrival, secondary dispersals between different European regions have been probably facilitated by shipping or oysters' transplants. In this sense, Garci et al. (2006) comment that *L. securis* could be unintentionally introduced in Galician waters via the importation of oyster seed from the

Mediterranean to shellfish farming areas; whereas Giusti et al. (2008) suggest the ballast water as the most likely vector of introduction of *L. securis* in the Central Tyrrhenian Sea.

Supporting this hypothesis, in a recent study, which included genetic identification of mussel samples collected from introduced and endemic areas, Pascual et al. (2010) suggest two historical introductions into European waters. One of these invasions seems to have started in Galicia, moving from there towards Italy and France; while the geographical spread of the second invasion cannot be deciphered, although a Australian/Pacific origin seems very possible.

In the Nervión estuary there is no knowledge concerning the arrival of *L. securis*. This estuary is connected with the sea through the commercial Port of Bilbao, one of the most important transport and logistics centres in the European Atlantic coasts. The absence of aquaculture activities along the Basque coasts suggests that naval transport is the most possible vector, either as larvae in ballast tanks or as adults attached to hulls.

Our observations, carried out on 20 February 2011, allows confirmation of the success of this invasion in the Ria of Nervión. Hundreds of dense populations and thousands of live and dead specimens colonize the inner part of Nervión estuary; revealing that the species is well established in the area.

The successful establishment of this species on the Basque coast may be facilitated by the thermal characteristics of ocean waters in the SE of Bay of Biscay. The fact that the surface waters are colder than the surrounding areas in summer months (Fischer-Piette 1935) has made possible the presence of a major number of southern species compared with proximal areas (Fischer-Piette 1938; Ibáñez 1978, 1985, 1989; Ibáñez et al. 1980, 1986). This reason could explain, partly, the successful establishment of a great number of native species from tropical and subtropical regions of Africa, North America, Asia and Australia (Martinez and Adarraga 2005b, 2006a).

On the other hand, *L. securis* has ecological traits that indicate that it may become highly successful in estuaries and semi-enclosed bays of the Bay of Biscay. These traits include: a short generation, gregariousness, abundance in its native habitat, wide environmental tolerance, ability to rapidly colonise new or disturbed substrata, capacity to colonise a wide range of

artificial structures and a relatively wide vertical tidal distribution. These characteristics often attributed to successful invaders (Ehrlich 1989; Fletcher and Farrell 1999) have facilitated the spread of this species in many habitats with significant environmental fluctuations.

As already mentioned, the first record outside its native range was in Kojima Bay, Okayama. Once established, it spread along the coast of Kanto, Chubu districts, Lake Hamana, and other sites in the south and west of Japan (Iwasaki 2004; Kimura et al. 2004). The average rate of spread for *L. securis* in Japan (only data available) has been estimated in 23.9 km/year (Iwasaki 2004). In the new habitats this mytilid can predominate among fouling organisms and benthos on tidal areas (Kajihara et al. 1976; Abdel-Razek et al. 1993a, b; Kimura 1994). Surveys on temporal patterns of abundance of *L. securis* in Lake Hamana, have showed two annual cohorts, in some areas where temperature and salinity fluctuate between 6.9°C–33.6°C and 2.6–31.3, respectively (Kimura and Sekiguchi 2008).

In the Mediterranean areas, this mussel habitually appears in association with *Mytilus galloprovincialis*, attached with the byssus on hard substrata or oyster shells in numerous colonies. The species is found exclusively in brackish waters and can tolerate broad variations in salinity (1 to 31) (Zenetos et al. 2003).

In Galicia, *L. securis* has colonised the upper reaches of estuaries in the inner part of the Ria de Vigo, where can tolerate salinities of between 5 and 36. The species forms numerous colonies in intertidal and shallow waters attached to rocks, artificial structures, or directly on muddy bottoms (Garci et al. 2006).

In the Nervión estuary the behaviour of *L. securis* is similar to the already mentioned areas. All specimens collected were found in the inner part. The salinity during a tidal cycle can fluctuate between 1.60 and 31; and the temperature between 10.6°C and 22°C. Bottom waters are hypoxic most of the time, and the organic matter contents in sediments are relatively elevated, around of 10% (Borja et al. 1996, 2006). The species appears forming regular colonies on all kind of natural and artificial substrata, on both soft and hard substrates.

Regarding the environmental effects in the Nervión estuary, it is too early to assess the real impact of this invader. At first, the establishment of this species could have a beneficial effect.

Pioneer colonization of mud and hard substrata could increase benthic macroinvertebrate abundance by providing habitat structure and bio-deposits (Ricciardi et al. 1997). However, an excess of stocks will inevitably have negative consequences. Due to the high filtration rates of mytilids [200–300 ml/min. (Bayne et al. 1976)], it is possible that suspension feeding by dense *L. securis* populations would affect to nutrient cycling, limiting food available to larvae of different organisms. On the other hand, the net respiration of a dense mussel bed may cause severe reductions in dissolved oxygen in flowing waters (Effler and Siegfried 1994). Other aspects that could be affected would be the turbidity levels, sedimentation, and pollutant transference. As mussels rapidly bioaccumulate contaminants, they could transfer metals and organochlorines to higher trophic levels (Kock and Bowmer 1993, MacIsaac 1996). Furthermore, given that *L. securis* can be attached to other molluscs (Garci et al. 2006), it may reduce native mussel populations by intense fouling similar to that caused by *Dreissena polymorpha* (Pallas, 1771) (Ricciardi et al. 1995, 1996, 1998). In terms of these impacts, Garci et al. (2006) mentioned the settlement of *L. securis* on the native mussel *M. galloprovincialis* in the Ria de Vigo; as well as a possible vertical displacement by *L. securis* of *M. galloprovincialis* to the intertidal upper rocky substrata. This competition with native species will also in all likelihood occur in the Nervión estuary. It is probable that, *M. galloprovincialis* will dominate the outer parts of the estuary, while *L. securis* will be dominant in the innermost ones. In the middle parts, strong competition for space with native fauna will take place.

Finally, given that *L. securis* is considered to be one of the worst invasive alien species in Europe (SEBI2010), further monitoring will be necessary in order to analyze the potential economic and ecological impact of *L. securis* invasion in this area.

## Acknowledgements

We would like to thank Ángel Borja, Iñigo Muxika and Javier Franco (AZTI-TECNALIA) and the Consorcio de Aguas de Bilbao-Bizkaia for providing subtidal samples; we also thank to Santiago Pascual, Manuel Garci (Instituto de Investigaciones Marinas ECOBIOMAR) and Michele Barbieri (Università di Pisa) for providing bibliography, and Olatz Adarraga for correcting the English of the text. Thank you to anonymous reviewers and the scientific editor for their constructive comments on the manuscript.

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**Supplementary material**

The following supplementary material is available for this article.

**Appendix 1.** Species composition and abundance of biota found in the four sampling stations where *Limnoperna securis* was collected.

This material is available as part of online article from:

[http://www.aquaticinvasions.net/2012/Supplements/AI\\_2012\\_2\\_Adarraga\\_Martinez\\_Supplement.pdf](http://www.aquaticinvasions.net/2012/Supplements/AI_2012_2_Adarraga_Martinez_Supplement.pdf)