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Research Article

First European record of the invasive barnacle *Balanus glandula* Darwin, 1854

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Abstract

During recent surveys of hard substrata biofouling communities in Belgian marine waters we discovered specimens of the invasive barnacle *Balanus glandula*. This species is new to the European marine fauna. The species was first encountered in July 2015 in a biofouling community on RV Belgica. In October 2016, the species proved to be common on navigational buoys in Belgian coastal waters and after a dedicated search we discovered the species on many groynes all along the Belgian coast and in the harbour of Zeebrugge. We identified the species both morphologically and genetically. We found two generations indicating that *B. glandula* had settled in 2015 (possibly the first year of its presence) and also in 2016. We provide identification characters to distinguish *B. glandula* from other, similar looking, Western European intertidal barnacles. The distribution of *B. glandula* was originally limited to the Pacific coast of North America. In the past half century *B. glandula* successfully invaded subsequently the coasts of Argentina, Japan and South Africa. Given its invasion history elsewhere, we forecast that *B. glandula* is on the brink of invading other European shores.

Key words: Belgium, invasive species, Cirripedia, introduced species

Introduction

Barnacles, as principal members of the marine fouling community, are amongst the most successful invaders and many species have been translocated all over the world, probably since the dawn of maritime transport. Chances for successful introductions elsewhere have accelerated for many barnacle species during the past decades (Torres et al. 2011). Shipping activities have increased in both numbers and speed, considerably shortening the voyage time and so increasing the likelihood for species to survive their journey to ultimately colonise areas beyond their natural range. Moreover, migrant barnacles now find more suitable habitat for settlement and growth as the number of artificial habitats in coastal areas and beyond steadily increases worldwide (e.g. Mineur et al. 2012).

A well-known example of a barnacle introduction is the post-WW II introduction of *Elminius* (*Austrominius*) *modestus* (Darwin, 1854) from New Zealand to Western Europe where it is now one of the most common intertidal barnacles (Bishop 1947; Crisp 1958; Harms 1999). More recently *Megabalanus coccopoma* (Darwin, 1845), originally limited to the tropical Pacific coast from Northern Baja California to the Gulf of Guayaquil (Newman and McConnaughey 1987), has colonized Brazilian waters, Louisiana, the North Sea, and lastly Japanese and tropical west African waters (Young 1994; Kerckhof and Cattrijsse 2001; Perreault 2004; Yamaguchi et al. 2009; Kerckhof et al. 2010).

Given its invasion history elsewhere (Geller et al. 2008) and the climatic suitability of north-western European marine waters, it was anticipated that *B. glandula* could be a candidate to invade Western European shores and, subsequently, the barnacle was recently suggested as a species of concern in a European horizon scanning exercise to derive a ranked list of alien species, which are likely to arrive, establish, spread and have an impact on biodiversity

Table 1. Overview of the number of features searched for (first number) and the presence of *Balanus glandula* (second number) along the Belgian coast.

	Groynes	Harbour walls	Buoys	Wind turbines	Belgica	Zeebrugge harbour	Oostende harbour
2013	4/0	0/0	30/0	4/0	1/0	0/0	0/0
2014	4/0	0/0	33/0	4/0	0/0	1/0	0/0
2015	6/0	0/0	24/0	2/0	1/1	1/0	0/0
2016	15/15	2/2	22/14	2/0	0/0	1/1	0/0
2017	9/9	2/2	12/10	–	–	–	–

or related ecosystem services in the European Union over the next decade (Roy et al. 2015). *Balanus glandula* was not, however, included in the final list of 95 species for which a risk assessment was considered a high to very high priority.

Balanus glandula Darwin, 1854 is a sessile (acorn) barnacle native to the Pacific coast of North America ranging from the Aleutian Islands to Bahia de San Quintín (Baja California) (Darwin 1854; Pilsbry 1916; Barnes and Barnes 1956; Newman 2007). During the past half century *B. glandula* has successfully invaded the coasts of Argentina (late 1960ies – first record 1974 (Spivak and L'Hoste 1976; Spivak and Schwindt 2014)) from San Clemente del Tuyú (Schwindt 2007) south to the Port of Ushuaia (personal observations, December 2016 where is was absent in 2005 (Schwindt 2007), the rocky shores along the west coast of Japan (Kado 2003) and, finally, over 400 km of coastline along the cooler upwelling areas along the west coast of South Africa (Simon-Blecher et al. 2008; Laird and Griffiths 2008). In South-Africa it has recently extended its range by 150 km by further invading the warmer South Coast (False Bay) thereby, and contrary to expectations, breaching the biogeographic break of the Cape Point (Robinson et al. 2015). In its native and also introduced ranges *B. glandula* is a common rocky intertidal barnacle, found in the middle to high intertidal zone.

This paper reports on the recent successful introduction of *B. glandula* into European waters.

Material and methods

Sampling

Focusing on non-native species we took samples of the biofouling community on various artificial and natural hard substrata in Belgian marine waters. The sampling is part of ongoing monitoring programmes that had recently been formalised in fulfilment of the requirements of the Marine Strategy Framework Directive of the European Commission.

Fouling organisms were collected qualitatively by scraping a certain surface area of the substratum or by performing searches for the presence of introduced species. The main artificial hard substrata searched for the presence of non-indigenous species were navigational buoys and various man-made structures along the Belgian coast such as groynes and harbour walls, offshore wind turbines and the two Belgian commercial coastal harbours.

Groynes are typically artificial hard substrate structures perpendicular to the beach meant to reduce beach erosion. During the past decades their numbers have increased and they are now the most common man-made structure on Belgian beaches. Navigational buoys deployed in Belgian marine waters typically lay out for two years after which they are landed for maintenance and cleaning. From 2000 on, every year, some 20 buoys distributed all over the Belgian part of the North Sea were searched upon landing for non-indigenous species. Additionally, we sampled the fouling of the Research Vessel (RV) Belgica (2012–2015) when in dry dock. The fouling—both intertidal and subtidal—on offshore wind turbines in Belgian waters has been investigated since 2008, immediately after the construction of the first structure (Table 1).

After the discovery of *B. glandula* on several navigational buoys in October 2016, we conducted a dedicated search for *B. glandula* on many groynes and harbour walls along the Belgian coast from December 2016 until March 2017. We measured the rostro-carinal diameter of intact individuals of *B. glandula* present in the samples from the buoys and groynes.

To assess the presence of reproductively mature individuals we examined, under a dissecting microscope, 40 selected individuals for the presence of brooded embryos *in casu* eyed nauplii. They were collected in March and early April 2017 on two groynes on the beach of Raversijde (Oostende). Since no gregarious populations or clusters were found, we sampled individuals living close (< 2 cm) to each other.

Selected specimens of *B glandula* were fixated in buffered formalin and afterwards transferred to denaturised ethanol for preservation. Additionally, a certain number of *B. glandula* were put directly in pure ethanol for genetic analysis.

We deposited voucher specimens in the Taxonomic Reference Collection of the Marine Ecology and Management (MARECO) team in Oostende and in the invertebrate collection of the Royal Belgian Institute of Natural Sciences in Brussels.

Identification

We identified the species morphologically using the classic publications by Darwin (1854) who first described the species, Pilsbry (1916) and Cornwall (1955).

Additionally, we performed a genetic analysis to confirm the identification (for details see Supplementary material Table S1). DNA was extracted from eight specimens, collected at four different locations along the Belgian coastal waters, with the Qiagen DNA Easy Blood and Tissue kit, according to the manufacture's protocol. Polymerase Chain Reaction (PCR) amplification (15 min at 95 °C, 40 cycles with 50 s at 95 °C, 50 s at 42 °C, 1 min at 72 °C, and a final extension step for 10 min at 72 °C) was performed on 500 bp of the cytochrome *c* oxidase I (COI) using the following primers: Forward primer LCO1490: 5'-GGTCAACAAATCATAAAGATATTGG-3' and Reverse primer HC02198: 5'-TAAACTTCAGG GTGACCAAAAAATCA-3' (Folmer et al. 1994). All amplifications were sequenced in both directions on an ABI 3130X automatic sequencer. Sequences are deposited in Genbank under accession numbers: BankIt2055448 Isolate1 MG214971 to BankItIsolate8 MG214978. Sequence similarity searches were performed using GenBank BLASTn (http://www.ncbi.nlm.nih.gov/BLAST/). Sequences were compared with COI sequences from the morphologically similar *Semibalanus balanoides* (Linnaeus, 1767) (18 specimens) and *Balanus crenatus* (Bruguiére, 1789) (15 specimens) collected in the North Sea (Raupach et al. 2015) and from *B. glandula* that were sampled at Fort Bragg in California (Sotka et al. 2004). At this site, the three haplotypes that could be distinguished within *B. glandula* were present. An overview of the accession numbers in GenBank is given in Table S2 in the supplementary material. Sequences were aligned and a 315 bp fragment was selected for further analysis. Intra- and interspecific nucleotide variability of the analysed barnacles was based on the Kimura 2-parameter model (Kimura 1980). A graphical representation was produced with neighbour joining cluster analyses (Saitou and Nei 1987), based on K2P distances using MEGA version 6 (Tamura et al. 2013). Nonparametric bootstrap support values were obtained by resampling and analysing 1,000 replicates (Felsenstein 1985).

Results

Our morphological and genetic analyses confirmed that we were dealing with *Balanus glandula*; i.e. our sequences returned a 100% match with *B. glandula*. When comparing with the COI sequences of *Balanus crenatus* and *Semibalanus balanoides*, the neighbour joining cluster based on the K2P distances showed no overlap between species clusters, with bootstrap values of 99% (Figure S1). The newly observed species for Belgium clusters with *B. glandula* that were sampled in their native region (Fort Bragg, California, USA).

Geographic spread in Belgian waters

We discovered the first *B. glandula* in 2015 (July 29th) when three small specimens were present in a large barnacle sample (constituting mainly of *Semibalanus balanoides*) taken from the highest zone, just below the waterline, on the hull of RV Belgica (Figure 2). The vessel had been operational since 30 January 2014, the previous dry docking and hull cleaning period. We had sampled the fouling of RV Belgica in the past on 17 December 2013 and on 19 July 2012, and detected no *B. glandula*. Accompanying barnacle species were: *Elminius* (*Austrominius*) *modestus, Semibalanus balanoides, Balanus crenatus*. Additionally, *Euraphia depressa* (Poli, 1791), *Balanus (Amphibalanus) improvisus* (Darwin, 1854), *Balanus trigonus, Solidobalanus fallax* (Broch, 1927) and *Balanus (Perforatus) perforatus* (Bruguière, 1789) were also present in the fouling community.

RV Belgica, based in the port of Zeebrugge (Belgium), mainly operates in the southern North Sea, mostly in Belgian waters, but every year, during summer, it makes a longer campaign to southern European waters. Hence, at the moment of discovery, the status of *B. glandula* in Belgian waters could not be ascertained.

Subsequently, from October 2016 on, with the exception of some off-shore buoys, we began to find *B. glandula* on almost every navigational buoy landed (Figure 1, Table S1). With the exception of several non-native barnacle species to the North Sea (Kerckhof et al. 2007), *B. glandula* was not detected on buoys between 2000 and 2015 and neither was the barnacle located on 5 other buoys inspected prior to October 2016.

We found specimens of *B. glandula* on every groyne searched between Oostduinkerke and Zeebrugge. The species was also recorded on the outer harbour walls of both Oostende and Zeebrugge and additionally also on structures in the latter port; *B. glandula* was not, however, found yet on any offshore wind turbines.

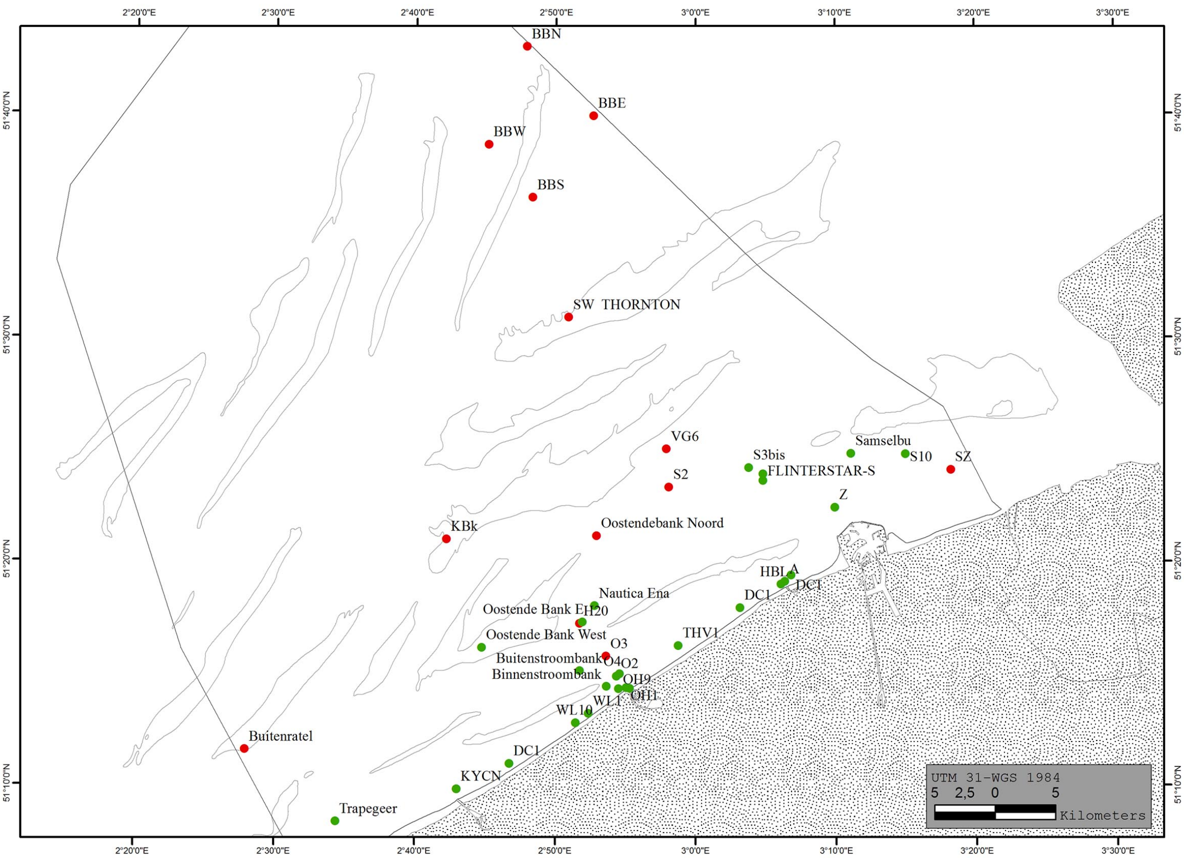

Figure 1. Map of the Belgian marine waters with the positions of the navigational buoys sampled for *Balanus glandula* since October 2016 red dots: absent, green dots: present.

Zonation and population features

On the buoys, *B. glandula* was only present in the highest, green algal zone, lying above the mussel belt. On the groynes, *B. glandula* lives in the high to middle intertidal zone and in both cases we found mostly solitary individuals, scattered over the substrate and not in gregarious populations or "clusters"; only rarely were the barnacles found in close association. Accompanying species were *S. balanoides* and *E. modestus*, whilst in the harbour of Zeebrugge *B. glandula* was also found in barnacle clusters together with *B. improvisus*. By the end of winter 2016–2017 the presence of *B. glandula* on the groynes became conspicuous as it was often the only barnacle left on an otherwise bare substrate (Figure 8). Those survivors were often severely eroded, but still alive.

The three *B. glandula* specimens from RV Belgica measured 5.39, 5.60 and 6.48 mm in rostro-carinal (R-C) diameter. On the groynes we found species up to 19.6 mm (groyne Wenduine, 12 December 2016) and from the buoys individuals reaching 23.8 mm in R-C diameter (buoy Z, in front of Zeebrugge, 20 March 2017).

None of the solitary settled individuals checked in March and April 2017 were brooding embryos although 33% of the specimens living in a radius of 2 cm of each other and collected in these months had eyed nauplii.

Discussion

Identification

Balanus glandula is a small white barnacle, typically measuring between 10–18 mm in R-C diameter, yet reaching to 22 mm (exceptionally: 30 mm) (Cornwall 1955; Newman 2007; personal observations) with six wall plates and a calcareous basal plate. As initially observed by Darwin, (who described the species in 1854), and later by Pilsbry (1916), *B. glandula* is extremely plastic displaying a variety of forms. Externally the shell surface ranges, as in *B. crenatus*, from strongly ribbed to smooth. The shape can be

Figure 2. *Balanus glandula*: external view of young specimen collected on the hull of RV Belgica July 29th 2015 Note sub-central dark area of each scutum (Photo: Cédric d'Udekem d'Acoz and Francis Kerckhof).

Figure 3. *Balanus glandula*: external view of ribbed conical specimen (Photo; Yves Barette).

Figure 4. *Balanus glandula*: external view of larger specimen (Photo: Yves Barette).

conic or more convexly, with solitary individuals tending to be more conic. In our material, most specimens display at least some ribbing although some smooth individuals were also present (Figures 2–4).

In general, the appearance of *B. glandula* may resemble several other local white barnacle species occurring in the intertidal zone in north Western Europe and hence it may be challenging to distinguish *B. glandula* from other local barnacles, especially in mixed and crowded populations. Therefore, to help identification, we provide an overview of a number of distinguishing characteristics visible in living specimens and empty tests of common intertidal white species (both native, cryptogenic and introduced) with which *B. glandula* can co-occur in north Western Europe (Table 2).

A characteristic feature of *B. glandula* is the presence, in the centre of the scuta, of a dark patch (Figure 2, 3 and 4), where the black inner surface of the tissue lining the terga and scuta shows through to the outside because the inner surface of the scuta is locally excavated into a deep pit (Pilsbry 1916). This enables the black colour of the underlying tissue to show through as a dark triangular spot. In cleaned scuta viewed with transmitted light, this central part of the scuta is translucent (Kado 2003) (Figures 5 and 6 herein). The black region is in particular visible in small individuals, although not always conspicuous in larger ones, especially when the barnacles are crowded or overgrown by algae. In certain populations the

Table 2. Comparison of selected characteristics for the identification of common white intertidal barnacles in north western Europe. Unique features in bold.

	Balanus glandula	*Balanus crenatus*	*Semibalanus balanoides*	*Elminius modestus*	*Balanus improvisus*	*Chthamalus* sp.	*Balanus balanus*
number of wall plates	6	6	6	**4**	6	6	6
basal plate or basis	calcareous, not permeated with tubes	calcareous, not permeated with tubes	membranous	membranous	solid, permeated with horizontal tubes, forming radiating pattern	membranous	calcareous, not permeated by radial tubes but with lines radiating from the centre
wall plates	not permeated, **internal centripetal ridges** visible at base	permeated with longitudinal tubes and transverse septa	not permeated, no internal centripetal ridges at base	not permeated	permeated with longitudinal tubes with transverse septa	not permeated	permeated with longitudinal tubes without transverse septa
colour of tergo-scutal flaps	dark maroon brown lining	light brown to purple, yellow rim, varying from strong yellow to almost white	predominately white, patches of black near the rostral end, central spot (micropyle) brown	white, with brown marks at the pylorus and one or two blackish bands in the rostral half	white speckled with purple, crossed by three conspicuous **black bands** (five if the outer ones are included in the count	**blue (brilliant or dull)** with orange or yellow central spot (micropyle)	strong yellow
tergum beaked	no	no	no	no	no	no	**yes**
scutum with sub central black area	**yes**	no	no	some greyish colour	no	no	no
lateral depressor muscle pit	**Conspicuous, translucent**	not conspicuous	not conspicuous	not conspicuous	not conspicuous	not conspicuous	not conspicuous
position on the shore	high - low intertidal	lower intertidal, subtidal	high intertidal	intertidal	subtidal, lower intertidal	high - low intertidal	subtidal, lower intertidal

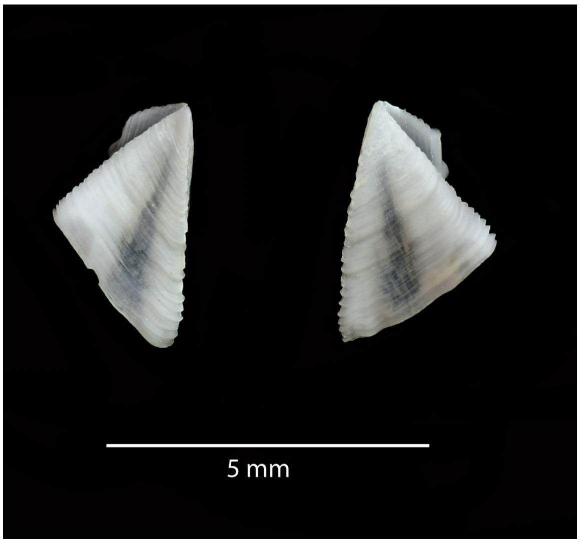

Figure 5. *Balanus glandula* scuta: external view showing the characteristic translucent black area. The darkness of the central region of each scutum is due to the black background showing through (see Figure 6), as the underlying hypodermis does *in vivo* (Figure 2) (Photo: Yves Barette).

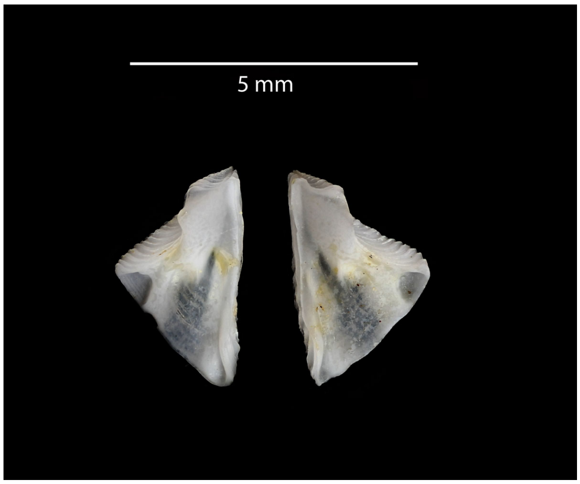

Figure 6. *Balanus glandula* scuta: internal view showing the very prominent articular ridge united with the very short adductor ridge and the small, marginal and deep depression for the lateral depressor muscle. The dark lateral depressor muscle pit as well as the central region of each scutum show up better than in Figure 5 because of the closeness of the thin areas in the plates to the black background (Photo: Yves Barette).

Figure 7. Comparison of internal basal view of the wall plates of, from the left to the right, *Semibalanus balanoides, Balanus glandula* and *Balanus crenatus*. The wall plates are not permeated by longitudinal tubes in *S .balanoides* and mature *B. glandula* in which centripetal ridges are visible. In *B. crenatus* the wall plates are permeated by longitudinal tubes with transverse septa. *Balanus crenatus* and *B. glandula* both have a calcareous basal plate or basis, here partly removed and partly visible (Photo: Thierry Hubin).

Figure 8. *Balanus glandula* in the high intertidal on a groyne, Raversijde, January 4th 2017. Left over basal plates of earlier dislodged individuals are also visible (Photo: Francis Kerckhof).

black zone is lacking so that other identification characters are required. *Balanus glandula* has a solid basal plate (calcareous basis) with a characteristic radiating pattern often left adhering to the substrate when the barnacle is removed. In young individuals, however, the calcareous basis might not be obvious when the barnacle is removed. The six wall plates are solid (except in some young individuals). Internally the wall plates are typically, strongly ribbed (Figure 7).

When dealing with living or fresh acorn barnacles the colour of the tissue lining the opercular plates on either side, the so-called tergo-scutal flaps, visible when the opercular plates are open, can be a very helpful character in distinguishing species (Southward 2008). These flaps are often white or strongly pigmented with a variety of colours including yellow, orange, red, brown, green, blue, purple and black and sometimes also display a pattern characteristic for a given species (see Table 2).

Internally the inside of the scutum of *B. glandula* (Figure 6) is characteristic, differing from all other species, as the articular ridge is very prominent and the adductor ridge very short, united above with the articular ridge and with a conspicuous pit as for a muscle below. The depression for the lateral depressor muscle is also small, marginal and deep.

Distinction between Balanus crenatus *and* Semibalanus balanoides

As noted by Darwin (1854), *B. glandula* closely resembles *B. crenatus* and *S. balanoides* particularly when small. The latter two species are very common intertidal barnacles along the northwestern European coasts and in the North Sea. Because *B. glandula* occupies a somewhat intermediate ecological position between *S. balanoides* and *B. crenatus* it is useful to consider the distinctions between the three species. *Balanus glandula* has, like *B. crenatus*, a calcareous basis, that is lacking in *S. balanoides*. The calcareous basis might not be obvious when individuals of *B. glandula* are removed and in young individuals it seems less firm than in similarly sized *B. crenatus*. The six wall plates of *B. crenatus* are provided with hollow longitudinal tubes (Figure 7), whilst those in *B. glandula* are solid (except in very young individuals). Internally the wall plates of *B. glandula* are typically strongly ribbed (Figure 7) and the tergo-scutal flaps are very dark, chestnut brown to even black, with no pattern unlike the tergo-scutal flaps of both *B. crenatus* and *S. balanoides,* which are never as dark.

Finally, in *B. glandula* the conspicuous pit on the inside of the scutum under the united short adductor ridge, the articular ridge and the conspicuous and deep marginal depression of the lateral depressor muscle (Figure 6) may be used to separate *B. glandula* from *B. crenatus* and *S. balanoides.*

First arrival in Europe

The presence of *B. glandula* specimens can easily be overlooked in the field, especially when overgrown and covered by algae and mud, or when crowded. Thus, the introduction of *B. glandula* to Japan, Argentina and South Africa initially remained unnoticed because of its resemblance to local species or because little attention was paid to the local intertidal fauna. In South Africa, *B. glandula* was first detected in 2007, but later proved to be present in 1992 or even earlier (Simon-Blecher et al. 2008). In Japan, *B. glandula* was discovered in 2000 but could have been introduced some 20–40 years earlier in the 1960s (Kado 2003). Coincidentally that was also the possible time that *B. glandula* invaded the Argentinian coast (Spivak and L'Hoste 1976).

The first Belgian specimens were collected end July 2015 from the hull of RV Belgica and given their size, we assume that they may have settled on the vessel in spring 2015 whilst in Belgian waters. A settlement of larvae in 2015 could also possibly be concluded from the presence of specimens of two size classes on several offshore buoys that were out at sea in 2015 and 2016 that were inspected in 2016, although, as this species may have multiple broods during a reproduction season, a multiple settlement in 2016 cannot be ruled out. In any case, larvae of the species must have been present in sufficient numbers to settle on different places along the Belgian coast in 2015 and the founder population must have been dense enough to ensure reproduction in Belgian waters by spring 2015.

We suspect that the inoculation could have happened in the harbour of Zeebrugge where the species is common. The port of Oostende, where the species is not yet present, is much smaller with less international traffic and no regular commercial shipping lines. Introductions in Japan and Argentina also occurred near large commercial ports. The initial 2015 settlement could have originated from larvae released from a fertile population present on the hull of a ship or larvae directly released from ballast water that settled *en masse* in and around the harbour of Zeebrugge, with individuals settling close enough to ensure a future reproduction.

Successful establishment

Balanus glandula can reproduce in its first year at a minimum reproductive size of 5.8 mm (Brown and Roughgarden 1985; Kado 2003; Leslie 2005). We found one year old fertile individuals in March and April 2017. In northern California *B. glandula* reproduces mostly in spring broods, but spawns from winter through late summer producing three to six relatively large broods (Hines 1978). As the larval development may last for four to eight weeks (Barnes and Barnes 1956; Hines 1978; Brown and Roughgarden 1985; Strathmann 1987) planktonic larval dispersal over distances of 100 km or more is possible; by the end of 2016, *B. glandula* was already widely spread along the Belgian coast. This rapid expansion is remarkable since no gregarious settling nor clusters were observed but rather scattered solitary individuals distributed all over the suitable substrate. Thus, a brooding population must have been present with at least some individuals close enough together to allow reproduction and the release of numerous nauplii.

The absence of any detected gregarious individuals raised the question of the reproductive capacities of the introduced population. Although in certain acorn barnacles self-fertilization is reported, it does not seem to occur in *B. glandula.* Kado (2003) found that solitary individuals were not fertile and thus did not contribute to the breeding population, an observation also made during our studies. Thus, to ensure reproduction, cross-fertilisation is needed with barnacles living close enough to each other. In certain populations, for example groynes around Raversijde, we regularly found individuals close enough to make cross-fertilisation possible although rarely touching each other. Wu (1981) found no fertilized individuals when barnacles were 5 cm apart but at 1.75 cm they were fertile. We found that 33% of individuals that were living in the 2 cm range of each other were fertile, similar findings to those of Kado (2003) who reported that 30% of the gregarious settled individuals were brooding. The recent discovery of spermcast mating in a stalked barnacle *Pollicipes polymerus* (Barazandeh et al. 2013), raises the possibility that eggs in the mantle cavity of isolated acorn barnacles could also be fertilized by sperm released by distant individuals. This was tested for *B. glandula* by Barazandeh et al. (2014) who found strong evidence that spermcast mating occurs, at least occasionally, in this species, although with lower incidence than reported for *P. polymerus*.

Possible impacts / effects

The arrival of *B. glandula*, in North Sea coastal waters marks the second incontestable invasion there of an acorn barnacle, the first being that of the Australian *Elminius (Austrominius) modestus* during World War II (Bishop 1947) since when this first invader has become established as a very common inhabitant of many North Atlantic shores (Crisp 1958). *Balanus glandula* now also contributes to the growing number of introduced species from around the world now present on Belgian intertidal artificial hard substrata (cf. Kerckhof et al. 2007 and De Mesel et al. 2015). In other areas where *B. glandula* was introduced, it has caused massive changes in intertidal biodiversity. Thus in Japan and South Africa it has out competed other barnacles, whilst in Argentina, where intertidal barnacles were previously absent (Darwin 1854), this invasive barnacle has outcompeted the native mussel *Brachidontes rodriguezi* (d'Orbigny, 1842) in the high intertidal of the exposed Mar del Plata coast (Vallarino and Elias 1997).

Along the Belgian coast, *B. glandula* occurs in the high and mid intertidal zone, where it co-occurs with *E. modestus* and *S. balanoides*, with which it is likely to compete. Competition with *B. crenatus* seems less likely since the latter lives lower in the intertidal zone. *Balanus glandula* is well adapted to survival in the highest intertidal zone as it is able to withstand desiccation over a long period (up to 13 days) (Carlton et al. 2011). Since *B. glandula* grows to a larger size and is more physically robust than native and already introduced European species such as *S. balanoides, E. modestus* and *Chthamalus* spp., we expect that this new invader will compete with other intertidal barnacle species now present and hence change the barnacle biodiversity and subsequently the overall biodiversity. This is expected to happen along the coasts of the southern North Sea and ultimately also along other north Western Atlantic European coasts. The introduced *E. modestus* population could also become affected as it is more fragile and remains much smaller although it produces multiple broods throughout the year. On the contrary the indigenous *S. balanoides* reproduces only once a year, in early spring. On the other hand large *B. glandula* may serve as a substratum for other sessile species; we have already found *B. glandula* that were completely covered by *E. modestus.* However, we don't expect that *B. glandula* will compete with the native blue mussel *Mytilus edulis* (Linnaeus, 1758) an important member of the intertidal fauna on hard substrata, because that species lives lower in the intertidal zone.

As to the possible origin of the Belgian population, this could be the native population in the northeast Pacific as well as from the other three introduced regions. Zeebrugge is a large international harbour with connections to ports all over the world, with several regular and important shipping lines to the Far East including the east coast of Japan.

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Supplementary material

The following supplementary material is available for this article:

Table S1. Presence/absence of *Balanus glandula* on navigational buoys since October 2016 and specimens selected for genetic analysis.
Table S2. Accession numbers of the sequences extracted from GenBank.
Figure S1. Neighbour joining tree based on K2P distances. BcrenNS: *Balanus crenatus* North Sea, SbalanNS: *Semibalanus balanoides* North Sea, BglanFB: *Balanus glandula* Fort Bragg (USA), BglanBE: *Balanus glandula* Belgium.

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