

**Research Article** 

# *Boccardia proboscidea* (Polychaete: Spionidae) in the SW Atlantic: how far has the invasion spread?

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## Abstract

*Boccardia proboscidea* is an exotic polychaete that was introduced to a Southwestern Atlantic Ocean coastal area of Argentina (Mar del Plata; Province of Buenos Aires). This polychaete proved to be a threat to local diversity as it displaced native species and modified the natural intertidal community structure. However, nothing is known about its latitudinal distribution in the country and the degree of its invasion. It is possible that due to deficiencies in taxonomy and lack of ecological studies, other localities of the Argentine coast were also invaded by this polychaete but have not yet been registered. The goal of the present study was to survey the latitudinal distribution of *B. proboscidea* in the coastal area of Argentina from 37°S to 54°S. In addition, this study aimed to evaluate the type of substrate colonized, investigate the presence of sewage effluence as a conditional factor for *B. proboscidea* establishment, and evaluate differences in the composition of species assemblages associated with the intertidal community invaded by the polychaete. *Boccardia proboscidea* was found latitudinally from 37°S to 47°S. The highest abundance was found on hard substrates and with intertidal sewage effluent. The opportunistic nature (r strategy) of *B. proboscidea* coupled with a continuous supply of organic matter (sewage effluent) may indicate the mechanism that has led to the success of its introduction into new localities.

Key words: latitudinal distribution, spatial survey, non-native species, polychaetes, intertidal fringe, sewage discharge, Argentina

#### Introduction

The global problem of biological invasions of exotic species and their impact is well known as a threat to local biodiversity (Ricciardi and Maclsaac 2000; Bax et al. 2003; Marques et al. 2013; Kerckhof and Faasse 2014). There are numerous records of benthic exotic species that have displaced native species and modified the natural community (Eno et al. 1997; Byers 2002; Bax et al. 2003; Richter 2010; Jaubet et al. 2013; Marques et al. 2013; Elías et al. 2015). The spionid polychaete *Boccardia proboscidea* (Hartman, 1940)

is native to California and has been introduced from British Columbia to Baja California (Hartman 1941; Hartman and Reish 1950; Berkeley and Berkeley 1950), Canada (Sato-Okoshi and Okoshi 1997), Hawaii (Bailey-Brock 2000), Argentina (Jaubet et al. 2011), Spain (Martínez et al. 2006), the North Sea (Kerckhof and Faasse 2014), the United Kingdom (Hatton and Pearce 2013), the English Channel (Spilmont et al. 2018), Japan (Imajima and Hartman 1964; Sato-Okoshi 2000), China Seas (Liu 2008), South Africa (Robinson et al. 2005; Simon et al. 2010), Australia (Blake and Kudenov 1978; Petch 1995; Hewitt et al. 2004), and New Zealand (Glasby et al. 2009). It is considered as an introduced species in all these countries and as an invasive species in many of them. Populations from South Africa are considered pests because they can cause mortality of important commercial species (Simon and Booth 2007; Simon et al. 2010). It has also been considered a potentially harmful species to oyster culture systems in Hawaii (Bailey-Brock 2000), Australia (Lleonart 2001) and New Zealand (Read 2004). In Argentina, B. proboscidea built biogenic reefs formed by 1,650,000 ind.m<sup>-2</sup> at a sewage-impacted site (Jaubet 2013). These reefs modified the structure of the natural intertidal community excluding the dominant ecosystem engineer, the mytilid Brachidontes rodriguezii (d'Orbigni, 1842) (Jaubet et al. 2013; Elías et al. 2015). To date, B. proboscidea has been registered at only 2 locations in Argentina: Puerto Madryn (42°49'46"S; 65°04'56"W; Diez et al. 2011) and Mar del Plata (38°00'S; 57°33'W; Jaubet et al. 2011), two port cities separated by a distance of 1200 km. However, the expansion of the B. proboscidea invasion in Argentina is as yet unknown and even well-established populations may remain undetected.

Spionids are found in a range of substrata and may form temporary or permanent tubes and burrows or live freely within the sediment. Some species also have the ability to bore into calcareous material (Hartman 1940; Blake 1981; Sato-Okoshi and Okoshi 1997; Simon et al. 2010; Hatton and Pearce 2013). Boccardia proboscidea has been reported at different habitat types/substrates including mudflats, sandy harbours, sandstone or sedimentary rocks, limestone reefs, and notably areas with sewage outfalls (Hartman 1940; Woodwick 1963; Imajima and Hartman 1964; Petch 1989; Gibson 1997). In addition, B. proboscidea is considered a good indicator of organic pollution due to its tolerance to high levels of organic matter. It is an opportunistic species in intertidal benthic communities (Johnson 1970) and has been found in sewage outfalls in Australia (Blake and Kudenov 1978), Spain (Martínez et al. 2006), and Argentina (Jaubet et al. 2011).

Reliable information on the presence (abundance/ density), habitat type, and latitudinal distribution of *B. proboscidea* are useful for understanding the invasion extent on Argentine shores and to predict/ prevent the serious consequences that can result in instability/imbalance of the invaded ecosystems. The aims of the work were to: 1) survey the latitudinal distribution of *B. proboscidea* in the coastal area of Argentina (from 37°S to 54°S); 2) evaluate the abundance of the polychaete and type of substrate colonized; 3) establish whether or not the presence of sewage effluence is a conditional factor for *B. proboscidea*  establishment and; 4) evaluate if there are differences in the species assemblages associated with the intertidal benthic community invaded by *B. proboscidea*.

# Material and methods

# Study area

The study area was located in the Southwestern Atlantic coastal area, along the central and south Argentine shores. The Argentinean marine coast is more than 4700 km long and includes twenty degrees of latitude (Miloslavich et al. 2016). Monitoring was carried out from 37° to 54° South latitude. The five provinces surveyed were Buenos Aires, Río Negro, Chubut, Santa Cruz, and Tierra del Fuego. The localities surveyed were Mar de Cobo, Santa Clara del Mar, Emisario Beach, Mar del Plata, Luna Roja, Miramar, Quequén, Pehuen-Co, Las Grutas, Puerto Madryn, Bahía Camarones, Comodoro Rivadavia, Puerto Deseado, Puerto San Julián, and Ushuaia (Figure 1 A and B, Supplementary material Table S1). In big cities like Mar del Plata, Puerto Madryn, and Ushuaia, more than one beach was sampled.

All intertidal zones sampled in the Province of Buenos Aires were characterized by beaches with rocky hard substrate, except Pehuen-Co which was characterized by sandy open beaches. Most intertidal platforms in this province are formed by consolidated sediments (stony rocks) that support both epilithic and endolithic biota (Amor et al. 1991; Bagur et al. 2013; Bagur et al. 2014). The Province of Rio Negro (Las Grutas) has intertidal platforms of varying substrate, including sedimentary (e.g., sandstones, limestones) and igneous rock types (e.g., granite, ignimbrites) (Kokot et al. 2004). The rocky shores of Chubut Province are dominated by a hard substrate of consolidated sediments. Puerto Madrvn and Comodoro Rivadavia cities are characterized by platforms of friable cineritic sediments, and Camarones city with platforms of sedimentary rocks although with some igneous rock (Miloslavich et al. 2016). The Province of Santa Cruz (Puerto Deseado and Puerto San Julián) is characterized by igneous rock substrates (Pankhurst et al. 1998). Finally, the Tierra del Fuego Islands (Bahia Ushuaia and Bahia Encerrada) are characterized by soft sand and mud bottoms.

At two localities in the study area (Quequén and Comodoro Rivadavia) sewage effluent was discharged directly into the intertidal zone (López Gappa et al. 1990; López Gappa et al. 1993; Mazón 2010). In both cases, the intertidal zones had sandy beaches interrupted by hard substrate (abrasion platforms). On the other hand, Emisario beach (in Mar del Plata city) is the site where the intertidal sewage effluence









**Figure 2.** Sampling sites invaded by *Boccardia proboscidea* at different latitudes in Argentina. The color of the circles represents the density range of *Boccardia proboscidea* (ind.m<sup>-2</sup>) at each sampling site. For details see Table S1.

formerly functioned. Currently at this site, the wastewater is discharged in the subtidal zone by mean of a submarine outfall 4 km from the coast (Cuello 2017; Cuello et al. 2017).

# Sampling design

Spatial monitoring was carried out in April 2016 and a total of 17 beaches were surveyed. Each sampling site was geographically located using GPS (Global Positioning System). The type of substrate (hard or soft) and the presence/absence of intertidal or subtidal sewage effluence were recorded at each beach. The types of substrate sampled were: sandy beaches, mud-sandy beaches, beaches with abrasion platforms (loessoides or cineritic sediments), beaches with outcrops of volcanic rocks (igneous rock), and beaches with sedimentary rocks.

In order to take samples of intertidal benthos at each site, between 7 and 10 independent sampling units were randomly collected. We followed the

354

methods of previous ecological studies (Vallarino 2013; Sánchez 2014; Elías et al. 2015). Benthic samples were taken by using a corer (PVC plastic cylinder of 10 cm diameter and 20 cm height; with an area of 78 cm<sup>2</sup>). The corer was buried into the community matrix until contact with the substrate (i.e down to the bottom). Samples were collected using a steel spatula placed between the corer and the substrate. The samples were preserved in 5% neutralized formalin solution. In the laboratory, each sample was sieved through a 0.5-mm mesh sieve and the retained organisms were identified, quantified, and preserved in 70% ethanol solution.

# Data analysis

A map of the distribution of *Boccardia proboscidea* along the Argentine coast was made integrating the monitored geographical points and the biological data (density) by means of a Geographic Information System (GIS) (Figure 2).

Generalized linear models (GLMs: Dobson 2002) were used to assess the distribution of *B. proboscidea* in relation to predictor variables. The explanatory variables evaluated were latitude, the presence or proximity of a sewage outfall, and type of substrate. Latitude is a continuous variable and both sewage outfall and type of substrate are categorical variables. Sewage outfall has three categories: with intertidal effluent (IE), with subtidal effluent (SE), and without effluence (WE); and substrate has four categories: loess/cineritic sediments (platforms); sand/mud; volcanic rock (VR), and sedimentary rock (SR). Only the main factors were evaluated: interactions between variables were excluded because of the high number of empty cells in the basic data matrix, due to the lack of data in the intersections between factors.

Because the occurrence of *Boccardia proboscidea* at each site was measured in 2 ways (presence/absence of the species and abundance), 2 types of generalized linear models were developed. First, to relate the presence or absence of *B. proboscidea* to habitat variables (latitude, sewage, and substrate), a logistic regression model with a binomial error structure and logit-link function was used (Venables and Dichmont 2004). Second, to relate the abundance of *B. proboscidea* to habitat variables, the model was fitted assuming a Poisson error distribution, with a log-link function. The standard errors of the parameter estimates and related statistics were computed taking into account over-dispersion (McCullagh and Nelder 1989).

The best model was selected using Akaike's Information Criterion (AIC; Akaike 1973). Models with a difference in AIC (dAIC) < 2 were considered to have equivalent support from the data (Burnham and Anderson 2002).

The community parameters (Species richness (S); evenness index (J') (Pielou 1969) and Shannon-Wiener diversity index (H') (Shannon and Wiener 1963)) were calculated for each sampling unit. The spatial variability of diversity parameters was evaluated compared to spatial variability of B. proboscidea abundance, and Pearson correlation analyzes were performed. Differences in the composition of the species assemblages relating to site (beach) were analyzed by combining a hierarchical agglomerative clustering using group complete linkage, and a non-metric multidimensional scaling (nMDS). PERMANOVA analysis on a Bray-Curtis similarity matrix after a 4th-root transformation was performed (Clarke and Warwick 2001). In this analysis, site was used as fixed factor. The SIMPER routine was used to determine the species accounting for the greatest contributions to the dissimilarity between assemblages. In order to aid understanding of generated plots (CLUSTER and nMDS) the sampling units for each sampling site were averaged. Vectors with the species that most contributed to the similarity between sampling sites were added to the nMDS according to the Pearson type correlation. The SIMPROF routine was used to assess whether the groups found in the cluster were significant or if they were obtained by chance.

# Results

We travelled a total of 2,945 km to conduct our research. *Boccardia proboscidea* was recorded in 13 of 17 coastal localities surveyed along the coastline of Argentina (70% of prevalence). This species was latitudinally distributed from the Province of Buenos Aires (37°S) to the Province of Santa Cruz (47°S). The invaded sites were Mar de Cobo, Santa Clara del Mar, Emisario Beach, Mar del Plata, Luna Roja, Miramar, Quequén, Pehuen-Co, Las Grutas, Puerto Madryn, Bahía Camarones, Comodoro Rivadavia, and Puerto Deseado (Figure 2, Table S1).

*Boccardia proboscidea* was found inhabiting different types of substrate: sandy, abrasion platform (loess and cineritic sediment), and volcanic rock. The mean density of *B. proboscidea* on hard substrate was 32,651 ind.m<sup>-2</sup> and on soft substrate was 27,768 ind.m<sup>-2</sup>. In turn, the density of *B. proboscidea* was significantly higher in sites with a contribution of organic matter (effluence). The mean densities were 118,718 ind.m<sup>-2</sup> in beaches with intertidal sewage effluence and 27,333 ind.m<sup>-2</sup> with subtidal sewage effluence.

In the presence/absence of B. proboscidea analysis, the model with the lowest value of AIC was model A that included latitude and the presence of sewage effluence (Table 1; AIC = 106.4). Both variables were statistically significant (Table 1). The predicted probability of occurrence of B. proboscidea decreased latitudinally from 37°S to 48°S (Figure 3A). On the other hand, the predicted probability of occurrence of *B. proboscidea* significantly increased at beaches with presence of intertidal effluence or submarine outfall (Figure 3B). In the abundance of B. proboscidea analysis, the models with the lowest value of AIC were models A and B (Table 2). These values had a difference of less than 2, therefore the simplest model was selected: model B that included only 2 variables. In this model, both the presence of sewage effluence and substrate were statistically significant (Table 2). The abundance of B. proboscidea increased significantly at beaches with a high contribution of organic matter (Figure 4A) and with abrasion platforms (loess and cineritic sediment) (Figure 4B).

The diversity indices were significantly different among sampling sites. Species richness (S) varied

**Table 1.** Results of the generalized linear models relating environmental variables (presence of a sewage effluence; type of substrate and latitude) to presence/absence of *Boccardia proboscidea*. GLM: Generalized linear model; df: degrees of freedom; AIC: Akaike information criterion; W: statistic Wald, p: significance level, \* indicates significant p values (< 0.05).

GLM					
presence/absence					
Distribution Binomial					
Model	df	AIC			
A-sewage+latitude	3	106.4532			
B-sewage+substrate+latitude	6	108.6276			
C-sewage+substrate	5	114.7254			
D-substrate+latitude	4	116.4700			
E-substrate	3	118.0659			
F-latitude	1	120.6009			
G-sewage	2	125.7540			
	df	Wald Stat.	р		
Intercept	1	14.0092	0.0001*		
Sewage	2	12.3181	0.0021*		
Latitude	1	12.3683	0.0004*		

**Table 2.** Results of the generalized linear models relating environmental variables (presence of a sewage effluence; type of substrate and latitude) to abundance of *Boccardia proboscidea*. GLM: Generalized linear model; df: degrees of freedom; AIC: Akaike information criterion; W: statistic Wald, p: significance level, \* indicates significant p values (< 0.05).

GLM					
abundance					
Distribution Poisson					
Model	df	AIC	2		
A-sewage+substrate+latitude	6	23118.10			
B-sewage+substrate	5	23118.45			
C-sewage+latitude	3	24347.68			
D-sewage	2	24431.02			
E-substrate+latitude	4	47192.74			
F-substrate	3	52103.71			
G-latitude	1	68576.43			
	df	Wald Stat.	р		
Intercept	1	724.831	0.000*		
Sewage	2	13809.708	0.000*		
Substrate	3	542.361	0.000*		



**Figure 3** Mean probability of occurrence of *Boccardia proboscidea* predicted by model at **A)** different latitude and **B)** with presence/ absence of intertidal/subtidal effluence. WE: without effluence; SE: subtidal effluence; IE: intertidal effluence.





between 2 to 15 species with highest richness values in Bahía Camarones (Chubut) and lowest values in Bahía Encerrada (Ushuaia) (Figure 5A). The evenness (J') and Shannon-Wiener diversity (H') index ranged from 0.32 to 0.80 and 0.43–1.86 respectively. The highest evenness and diversity values were found in San Julián (Santa Cruz), while the lowest values were found in Mar de Cobo (Buenos Aires) and Bahía Encerrada (Ushuaia) respectively (Figure 5B and C). However, no significant correlation was found between the spatial variability of *B. proboscidea* abundance ((S, *B. proboscidea*):  $r^2 = 0.031$ ; N = 96; p = 0.085; (J', *B. proboscidea*):  $r^2 = 0.032$ ; N = 96; p = 0.080; (H', *B. proboscidea*):  $r^2 = 0.039$ ; N = 96; p = 0.052)).

PERMANOVA showed that the structure of the benthic community was significantly different among sites (Pseudo-F<sub>site</sub> = 16.679; df = 15; p = 0.001). The

ranking of species that most contributed to the dissimilarity (cut to 50%) among the sampling sites are shown in Supplementary material Table S2. The sampling sites were separated significantly into three groups in the dendrogram clustering plot according to > 70% dissimilarity (SIMPROF with significance level = 5%; Group 1 (Pi = 1.92; p = 2.6); Group 2 (Pi = 3.96; p = 0.1) and Group 3 (Pi = 5.15; p = 0.1)(Figure 6A). A similar pattern was distinguished in the nMDS ordination plot (Figure 6B). All sampling sites were different, with the exception of the groups: "Mar del Plata-Miramar", "Mar de Cobo-Santa Clara-Emisario beach", and "Comodoro Rivadavia-Puerto Madryn", where those communities were not different (Similarity 60%). The species Boccardia proboscidea and Monocorophium insidiosum (Crawford, 1937) were correlated with beaches with hard substrate and with effluent discharge; while Brachidontes rodriguezii



Figure 5. Spatial variation of the abundance of *Boccardia proboscidea* and community parameters: A) Species richness; B) Evenness index and C) Shannon-Wiener diversity index at each sampling sites. MC: Mar de Cobo; E: Emisario Beach; SC: Santa Clara del Mar; MDP: Mar del Plata; LR: Luna Roja; M: Miramar; QQ: Quequén; LG: Las Grutas; PM: Puerto Madryn; PMA: Puerto Madryn sandy; BC: Bahía Camarones; CR: Comodoro Rivadavia; PD: Puerto Deseado; SJ: San Julián; BU: Bahía Ushuaia and BE: Bahía Encerrada.

and *Siphonaria lessoni* Blainville, 1827 were correlated with beaches with hard substrate and without effluent discharge. The Oligochaeta were correlated with beaches located at high latitudes and without effluence (Corr. Pearson = 0.75; n = 16; r<sup>2</sup> = 0.5625) (Figure 6B).

# Discussion

This study presents the first assessment of the latitudinal extent of the introduced *Boccardia proboscidea* polychaete in the SW Atlantic (Argentina). This study also provides a modelling analysis of occurrence and distribution of the species in relation



**Figure 6.** Differences in the species assemblages at the sampling sites combining: A) a hierarchical agglomerative clustering; and B) a non-metric multidimensional scaling (nMDS). Cluster formed were superimposed on the 2-dimensional nMDS obtained from the similarity matrixes. The species that most contributed to the similarity between sampling sites were added according to the Pearson type correlation (r = 0.75). MC: Mar del Cobo; E: Emisario Beach; SC: Santa Clara del Mar; MDP: Mar del Plata; LR: Luna Roja; M: Miramar; QQ: Quequén; LG: Las Grutas; PM: Puerto Madryn; PMA: Puerto Madryn sandy; BC: Bahía Camarones; CR: Comodoro Rivadavia; PD: Puerto Deseado; SJ: San Julián; BU: Bahía Ushuaia and BE: Bahía Encerrada.

to environmental variables (latitude, type of substrate, and presence of a sewage discharge). Only latitude and effluence presence significantly influenced their occurrence and abundance. This type of analysis can be useful for predicting/preventing future introductions of *B. proboscidea* in localities that have not yet been invaded in Argentina and elsewhere.

Boccardia proboscidea was originally described from the west coast of North America (California) (Hartman 1940) and has subsequently been recorded, with a wide geographical distribution, in the Pacific Ocean and to a lesser extent in the Atlantic Ocean (Jaubet et al. 2015). The species has been transported, presumably from the NE Pacific to distant locations, and now has a distribution that is nearly cosmopolitan (Blake et al., in press). Marine systems are particularly susceptible to biological invasions due to global travel and shipping (Bax et al. 2003). Shipping has contributed to 69% of all marine introductions and is considered the most common invasion pathway (by carrying invasive species in ballast or by biofouling) (Molnar et al. 2008). Ports can act as hotspots for invasive species due to heavy shipping traffic (Hewitt 2002). Boccardia proboscidea could have

been transported out of its native range by mean of vessels (ballast water, biofouling, etc.) or with the import of species of commercial interest for aquaculture (Carlton 1985; Carlton and Geller 1993; Bailey-Brock 2000; Ruiz et al. 2002; Carlton and Ruiz 2005; Diez et al. 2011; Simon et al. 2009; Simon et al. 2010). The transport of bivalve mollusks for commercial interest (such as oysters) from Japan to California has been a route for the introduction of a spionid polychaete (*Pseudopolydora*) that bores the shells of this mollusk (Blake and Woodwick 1975; Carlton 1975).

In Argentina, *B. proboscidea* was latitudinally found from 37°S to 47°S (present study). The possible route of entry for the species to the southwestern Atlantic Ocean ports could be increasing maritime traffic in recent times. The port of Puerto Madryn city could be a possible zone of introduction for larval, juvenile, or adult *B. proboscidea*. The deep water port is located next to an industrial plant for aluminum production. The imports of the raw material (bauxite) necessary for the production and subsequent exportation of produced metal (aluminum) come from Australia (http://www.cei.mrecic.gov.ar). This aluminum plant, located in Puerto Madryn, was installed in 1974 (http://www.aluar.com.ar). Interestingly, *B. proboscidea* was reported from Port Phillip Bay, Victoria, Australia, in 1978 (Blake and Kudenov 1978), reaching densities up to 164,000 ind.m<sup>-2</sup> in areas of secondary treated sewage discharge (Dorsey 1982). Therefore, these data supports the idea that the maritime connection between Australia and Argentina (Puerto Madryn) could have enabled, in the 1970s, the introduction of this species into Argentina.

On the other hand, the city of Quequén, located 4 km east of the port of Quequén (Province of Buenos Aires), and the city of Comodoro Rivadavia (Province of Chubut), located 1,760 km south of Buenos Aires, are sites with a high probability of finding presence/high abundance (i.e. establishment success) of B. proboscidea, since, in addition to being port cities, they have discharges of intertidal wastewater effluent. Several studies were conducted in the port of Ouequen in order to evaluate the response of the benthic community to the intertidal sewage discharge (López Gappa et al. 1990; López Gappa et al. 1993; Adami et al. 2004). However, these studies, carried out two decades ago, only mentioned the presence of dense infaunal populations of Boccardia sp., Boccardiella sp. (Blake and Kudenov 1978), and Boccardia polybranchia (Haswell 1885) in the highly polluted area. Boccardia proboscidea was recently recorded as the dominant species among the zoobenthos at the impacted site (areas surrounding the sewage discharge at Quequén), and as impeding the settlement of other species and drastically decreasing diversity parameters (Becherucci et al. 2018). In Comodoro Rivadavia, we expected to find a greater density of *B. proboscidea* in areas surrounding the sewage discharge. However, various species of the genus Boccardia were found; including Boccardia proboscidea, Boccardia polybranchia, and Boccardia sp. Interspecific competition for space and food could have played an important role at this locality. Therefore, B. proboscidea could have entered the coasts of the Argentine sea through the ports of Quequen, Puerto Madryn, or Comodoro Rivadavia. We hypothesize that the first introduction could have been in the south of the country (Puerto Madryn or Comodoro Rivadavia) and by means of coastal maritime traffic, the species could have been dispersed along the Argentine coast. Due to prevailing littoral south to north current, the spread was likely mostly to the north, while to the south there was likely less traffic and no current. An exhaustive taxonomic revision of all material deposited in museums of Argentina would be necessary to confirm the possible date and site of its initial introduction.

*Boccardia proboscidea* is an opportunistic species in intertidal benthic communities (Johnson 1970) and is considered an indicator of organic contamination (Petch 1989). The opportunistic nature of *B. proboscidea* includes tolerance for a wide range of habitats, including variable salinities and temperatures (Hartman 1940) and a larval biology that includes both lecithotrophic and planktotrophic larvae in the same populations (Blake and Kudenov 1981; Gibson 1997; Gibson and Gibson 2004; Gibson and Smith 2004; Blake et al., *in press*). The results obtained in this study showed that *B. proboscidea* colonized a variety of substrates but hard substrate was most colonized. Only two beaches with soft substrate (sand) were colonized (Pehuen-Co and Puerto Madryn). In Puerto Madryn, *B. proboscidea* was recorded inhabiting both stony and sandy beaches (Diez et al. 2011).

The highest values for *B. proboscidea* abundance were found at beaches with intertidal sewage effluence. The extra supply of organic matter probably provides polychaetes with abundant food, which could favor quick colonization of available space. Organic material acts as an environmental trigger and the populations of this opportunistic species may grow and spread over the intertidal benthic community more rapidly because of abundant nutrients. Previous studies carried out in the intertidal rocky shore at Mar del Plata showed the tolerance of *B. proboscidea* to organic pollution and the weakness of the bivalve Brachidontes rodriguezii, an ecosystem engineer that was competitively excluded by *B. proboscidea* at sewage-impacted areas (Jaubet et al. 2013; Elías et al. 2015; Garaffo et al. 2016; Becherucci et al. 2018). On the other hand, the abundance of *B. proboscidea* was also high at the beaches affected by current subtidal sewage effluence (Emisario Beach, Mar del Plata). Although this subtidal effluence was inaugurated in December 2014 and discharges of organic content are 4 km offshore, the construction period altered the sedimentary dynamics of the area (by the addition of 2 breakwaters), and subsequently the decrease in the organic matter levels of the sediment was slower than expected (Cuello 2017; Cuello et al. 2017). In addition, B. proboscidea reefs were formerly found on this site, with densities up to 1.6 million individuals per square meter (in both endolithic and epilithic forms) (Jaubet 2013). Although the discharge now occurs almost 4 km offshore, the intertidal rocks still contain large densities of this species.

At 49° South latitude, the abundance of *B. proboscidea*, even at port cities (Puerto San Julian and Ushuaia), was zero. Their absence could be related to the low temperature of the water in these latitudes (average values:  $5.1 \,^{\circ}$ C in winter and  $7.5 \,^{\circ}$ C in summer; Borla and Vereda 2015) or to the lack of an appropriate substrate (hard substrate) but perhaps mainly due to the absence of an intertidal sewage discharge. The lack of a littoral current from north to south and the little maritime traffic could also explain why *B. proboscidea* was not introduced or established at higher latitudes.

The species assemblages were different in each sampling site. The study area comprises two biogeographical provinces (Argentinean and Magellanic) delimited by the Valdés Peninsula. The Argentinean Biogeographic Province extends from 36° to 43°S latitude and includes the Provinces of Buenos Aires and Río Negro, and the north of Chubut Province. The Magellanic Biogeographic Province, extends from 43° to 56°S latitude along southern Chubut Province as well as Santa Cruz and Tierra del Fuego Provinces (López Gappa et al. 2006; Balech and Ehrlich 2008; Miloslavich et al. 2016). Therefore, the differences in the composition of the assemblage of species were attributed to the characteristics of each site. In general, when latitude increases, the type of substrate, the habitat, and the *B. proboscidea* benthic community structure changes.

## Conclusion

The latitudinal distribution of the invasive species Boccardia proboscidea in Argentina ranges from 37°S to 47°S. Various types of substrate were colonized by this species (sand, loess platform, platform of friable cineritic sediments and volcanic rock). The highest values of abundance were found on beaches with abrasion platforms (loess and cineritic sediment) and beaches affected by intertidal sewage effluents. Although the species was introduced from Mar de Cobo (37°S) to Puerto Deseado (47°S), it could be considered an invasive species in Ouequén (38°S) and Comodoro Rivadavia (45°S) due to the high abundance found and to the similarity in the environmental conditions with the site where the species developed biogenic reefs (in 2008, in Mar del Plata). The spatial monitoring carried out in this study of the beaches of Argentina invaded by *B. probosocidea* is useful for better understanding the extent of the invasion and to predict the possible places suitable for future introductions.

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#### Supplementary material

The following supplementary material is available for this article:

**Table S1.** Details of Argentina localities sampled during the spatial survey: site name, province, geographical coordinates and presence of *Boccardia proboscidea*.

**Table S2.** SIMPER results showing the similarity within each locality including only those species that contributing to 50% similarity and the dissimilarity between locations including the species that most contribute to this dissimilarity (cut to 50%).

This material is available as part of online article from: http://www.aquaticinvasions.net/2018/Supplements/AI\_2018\_Jaubet\_etal\_SupplementaryTables.xlsx