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Short communication

# Potentially toxic *Pseudo-nitzschia* species in plankton and fecal samples of *Eubalaena australis* from Península Valdés calving ground, Argentina

Valeria C. D'Agostino [a,*], Mónica S. Hoffmeyer [b,c], Gastón O. Almandoz [d,e], Viviana Sastre [f], Mariana Degrati [a,g]

[a] Centro Nacional Patagónico (CENPAT), Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET), Boulevard Brown 2915, U9120ACD Puerto Madryn, Argentina
[b] Instituto Argentino de Oceanografía (IADO), CONICET-Bahía Blanca, Camino La Carrindanga Km 7.5, B8000FWB Bahía Blanca, Argentina
[c] Facultad Regional Bahía Blanca, Universidad Tecnológica Nacional, 11 de Abril 461, B8000LMI Bahía Blanca, Argentina
[d] Facultad de Ciencias Naturales y Museo, Universidad Nacional de La Plata, Paseo del Bosque S/N, B1900FWA La Plata, Argentina
[e] Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET), Argentina
[f] Facultad de Ciencias Naturales, Universidad Nacional de la Patagonia San Juan Bosco, Roca 115, 1er Piso, U9100AQC Trelew, Argentina
[g] Universidad Nacional de la Patagonia San Juan Bosco, Boulevard Brown 3150, U9120ACV Puerto Madryn, Argentina

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ABSTRACT

Península Valdés (PV) in Argentina is an important calving ground for the southern right whale *Eubalaena australis*. However, a high mortality of calves has been observed in the last years, which could be associated with phycotoxin exposure. During a sampling program conducted late in the calving seasons of 2004, 2005 and 2010, potentially toxic species of the genus *Pseudo-nitzschia* were observed to be an important component of the phytoplankton community and they were also found in fecal samples of two live whales and three stranded whales. In line with this, in the present study *Pseudo-nitzschia australis, Pseudo-nitzschia fraudulenta, Pseudo-nitzschia pungens* and the complex *Pseudo-nitzschia pseudodelicatissima* were identified in fecal samples and phytoplankton samples by light and electron microscopy. Although no toxin analysis was carried out in the present study, our findings suggest that *E. australis* could be exposed to domoic acid in their calving ground.


## 1. Introduction

Some diatom species of the genus *Pseudo-nitzschia* produce domoic acid (DA), a neurotoxin that causes Amnesic Shellfish Poisoning (ASP). DA can be transferred along the marine food web to marine mammals via organisms that feed on the toxin-producer *Pseudo-nitzschia*. DA has therefore been associated with several mortality events among large marine mammals, including dolphins, sea lions and whales (Lefebvre et al., 1999; Gulland et al., 2002; De la Riva et al., 2009).

Península Valdés (PV) is an important calving ground for the southern right whale (*Eubalaena australis*) population in the south-western Atlantic Ocean. In recent years (2007–2011) an increase in the number of calves that were found dead on the shores of PV was recorded (Rowntree et al., 2013). At least four hypotheses were proposed to explain the possible causes of death, namely: a) decrease in food abundance, b) exposure to biotoxins produced during harmful algal blooms, c) infectious diseases, and d) effects derived from kelp gull harassment (Rowntree et al., 2013). *E. australis* is typically found in Nuevo Gulf (NG) and San José Gulf (SJG), two surrounding gulfs in PV, from May to December, evidencing peak numbers during August–September (Crespo et al., 2014). During their stay in PV adults and juveniles begin to feed sporadically in late September–early October, when denser zooplankton patches follow the spring phytoplankton blooms (Payne, 1995; Sironi, 2004; Hoffmeyer et al., 2010).

The following potentially toxic *Pseudo-nitzschia* species have been reported to be present in NG and SJG: *P. australis*, *Pseudo-nitzschia calliantha, Pseudo-nitzschia delicatissima*, *P. fraudulenta*, *Pseudo-nitzschia multiseries* and *P. pungens* (Sastre et al., 2001; Cadaillón, 2012). The presence of DA was detected for the first time in phytoplankton samples of NG in October 2005 in a concentration of 0.66 μg/100 mL during a spring phytoplankton bloom dominated by *P. pungens* and *P. fraudulenta* (Sastre et al., 2007). More recently, Cadaillón (2012) determined the presence of DA in phytoplankton and zooplankton in a study carried out in both NG and SJG. In this study, DA concentrations were observed to range from 0.75 to 40.96 μg DA $g^{-1}$ in phytoplankton samples and from 0.43 to 25.75 μg DA $g^{-1}$ in zooplankton samples from NG, whereas they were found to range from below the detection limit to 4.88 μg DA $g^{-1}$ in phytoplankton samples and from 2.55 to 42.78 μg DA $g^{-1}$ in zooplankton samples from SJG. This suggests the role of zooplankton as a potential vector for DA and the possible toxin transfer to higher levels of the food web, thus being a potential risk for *E. australis* as well as for other animals that usually forage on zooplankton.

* Corresponding author at: Centro Nacional Patagónico (CENPAT), Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET), Boulevard Brown 2915, U9120ACD Puerto Madryn, Argentina.
*E-mail address:* valeriacdagostino@gmail.com (V.C. D'Agostino).

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Previous research conducted in the northern hemisphere detected DA and frustule fragments of *Pseudo-nitzschia* spp. in fecal samples of stranded and live individuals of the North Atlantic right whales (*Eubalaena glacialis*) that had been feeding in their feeding grounds in spring and summer (Leandro et al., 2010). DA levels in zooplankton samples (mostly copepods) collected during their study ranged from 0.02 to 0.17 μg DA $g^{-1}$. In parallel, the presence of remains of the copepod *Calanus finmarchicus* in all whale feces led to the conclusion that *E. glacialis* was exposed to DA by ingestion of contaminated copepods (Leandro et al., 2010). Similarly, trace levels of saxitoxin and DA were detected in samples of feces, urine and tissues collected from dead individuals of *E. australis* in PV (Uhart et al., 2009; Rowntree et al., 2013). On account of the fact that the presence of toxic algae in fecal samples from these whales is an issue that remains unexplored to date, the purpose of the present study was to examine fecal samples from *E. australis* collected from their calving ground in PV in order to trace the presence of *Pseudo-nitzschia* species. Phytoplankton samples from the same area were also analyzed to compare their composition with fecal content.

## 2. Material and methods

### 2.1. Sampling of fecal and phytoplankton samples

Two fecal samples from live whales were collected from an area close to Puerto Pirámides (NG) (Fig. 1) using a 300 μm mesh net attached to a stick by personnel working for commercial whale-watching boats in the spring of 2004 and 2005. Samples were preserved in 70% alcohol. Each of these fecal samples, which were compact material that floated together after defecation, belonged to one whale. Whale feces from three stranded individuals from SJG beaches were collected by members of the Southern Right Whale Health Monitoring Program (SRWHMP) during the spring of 2010 (Fig. 1). These samples were directly collected from the intestine from each individual during necropsy. Samples were preserved in neutralized formalin 5%.

Phytoplankton samples were collected in the frame of the Harmful Algae Monitoring Program (Ministry of Fisheries — Chubut Province, Argentina) on similar dates from sites close to those where whales' feces had been collected (Fig. 1, Table 1). Samples were collected with a Van Dorn bottle and fixed with Lugol solution for quantitative analysis. Additional samples were collected using a 25 μm mesh and they were fixed with formaldehyde 4% for qualitative analysis.

### 2.2. Laboratory procedures and analyses

Whale feces were examined by scanning electron microscope (SEM) (Jeol JSM-6460 LV) for *Pseudo-nitzschia* spp. identification following Leandro et al. (2010) with slight modifications. One mL of each fecal sample was pipetted onto 3 μm pore, 47 mm diameter polycarbonate filter and rinsed under low vacuum with 10 mL of distilled water. Samples were subsequently dehydrated with 4 mL of a series of acetone (10% to 80%) followed by 8 mL of 100% acetone. Filters were air-dried for ~5 min and finally placed onto double-sided tape glued on glass slides. Samples were sputter-coated with gold before examination.

Phytoplankton was quantified with an inverted microscope following Lund et al. (1958). Cleaning of *Pseudo-nitzschia* spp. frustules was performed according to the method of Hasle and Fryxell (1970). Naphrax mounted slides were observed with an optical microscope equipped with phase contrast and selected samples were observed by SEM (Jeol JSM-6360 LV) for species identification.

## 3. Results and discussion

Frustule fragments of *Pseudo-nitzschia* spp. were found in all the fecal samples analyzed (Table 1). Similarly, the genus *Pseudo-nitzschia* was detected in all the phytoplankton samples analyzed except in those collected from Pardelas Beach (NG) on 24 October 2004. At least four taxa were identified in the fecal samples and phytoplankton samples analyzed, namely *P. australis*, *P. fraudulenta*, *P. pungens* and the complex *P. pseudodelicatissima* (Fig. 2). The first three species are worldwide known as DA producers (Lelong et al., 2012) and are also

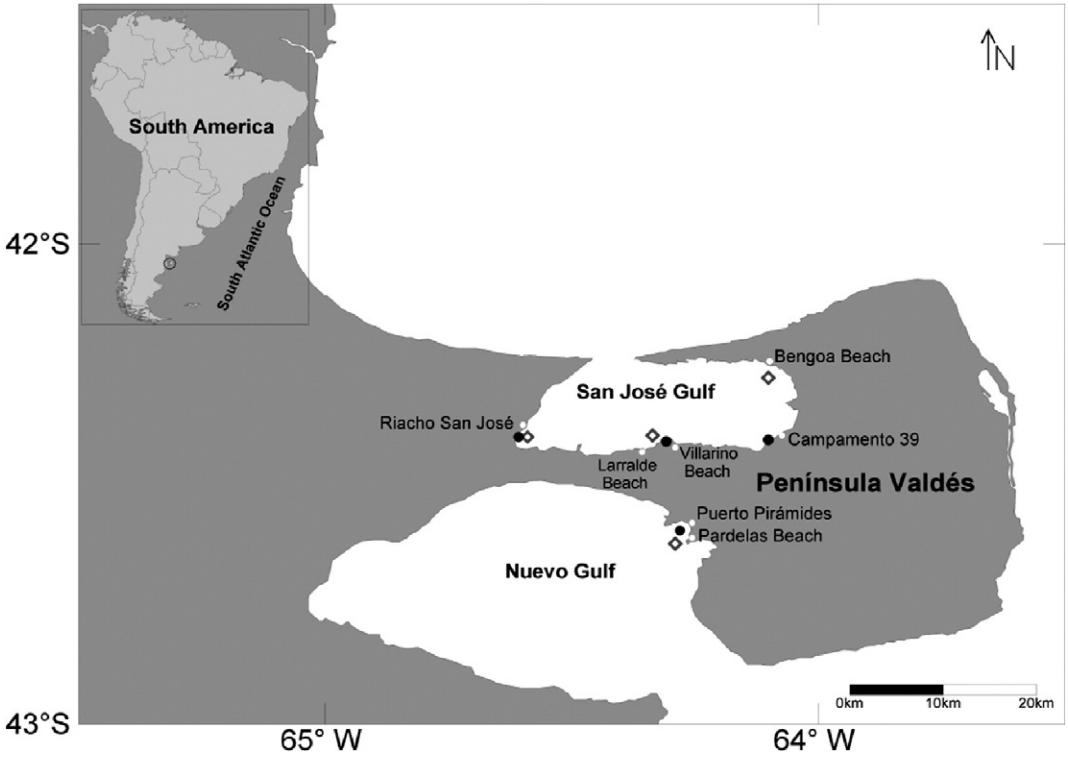


**Fig. 1.** Península Valdés, Argentina. Collection sites of whale fecal (•) and phytoplankton samples (◊).

**Table 1**
*Pseudo-nitzschia* spp. present in *Eubalaena australis* fecal samples of live and stranded whales in Nuevo Gulf (NG) and San José Gulf (SJG) respectively, and their density in phytoplankton samples collected in the area where fecal samples were found.

| Sample | Sample collection date | Sample location | Lat. (°S) | Long (°W) | *Pseudo-nitzschia* spp. | Abundance (cell $L^{-1}$) |
|---|---|---|---|---|---|---|
| Live whale feces (n-i) | October 2004 | Puerto Pirámides (NG) | 42°36.2′ | 64°19.9′ | *P. pungens* | n-c |
| Phytoplankton | 10/24/2004 | Pardelas Beach (NG) | 42°37.4′ | 64°16.0′ | n-d | n-d |
| Phytoplankton | 9/13/2005 | Pardelas Beach (NG) | 42°37.4′ | 64°16.0′ | complex *P. pseudodelicatissima* | 2200 |
| Live whale feces (n-i) | 09/26/2005 | Puerto Pirámides (NG) | 42°36.2′ | 64°19.9′ | *P. pungens* | n-c |
| Phytoplankton | 09/09/2010 | Larralde Beach (SJG) | 42°24.2′ | 64°18.9′ | *P. pungens* | 30,240 |
| | | | | | complex *P. pseudodelicatissima* | 26,880 |
| Stranded whale feces (adult female) | 09/14/2010 | Villarino Beach (SJG) | 42°25.0′ | 64°18.0′ | *P. pungens* | n-c |
| Phytoplankton | 10/14/2010 | Riacho San José (SJG) | 42°24.1′ | 64°36.5′ | *P. pungens* | 8400 |
| | | | | | complex *P. pseudodelicatissima* | 16,800 |
| | | | | | *P. fraudulenta* | 183,120 |
| Stranded whale feces (juvenile female) | 10/27/2010 | Riacho San José (SJG) | 42°23.2′ | 64°36.0′ | complex *P. pseudodelicatissima*; *P. pungens* | n-c |
| Stranded whale feces (adult female) | 11/09/2010 | Campamento 39 (SJG) | 42°22.4′ | 64°6.0′ | complex *P. pseudodelicatissima*; *P. australis* | n-c |
| Phytoplankton | 11/11/2010 | Larralde Beach (SJG) | 42°24.2′ | 64°18.9′ | *P. fraudulenta* | 1,001,280 |
| | | | | | complex *P. pseudodelicatissima* | 1,162,560 |
| Phytoplankton | 11/11/2010 | Bengoa Beach (SJG) | 42°15.4′ | 64°6.2′ | *P. fraudulenta* | 1,327,200 |
| | | | | | complex *P. pseudodelicatissima* | 275,520 |

NG: Nuevo Gulf; SJG: San José Gulf; n-c: not counted; n-d: undetected; n-i: unidentified developmental stage and sex.

associated with toxic events in Argentinean waters (Negri et al., 2004; Sastre et al., 2007). On the other hand, both toxic and non-toxic species are included in the complex *P. pseudodelicatissima*, which contains several cryptic and pseudocryptic species that can only be identified by means of molecular tools (e.g. Lelong et al., 2012). Interestingly, other toxic (i.e. *P. turgidula*) and non-toxic *Pseudo-nitzschia* species (e.g. *P. lineola*, *P. turgiduloides*, *P. subcurvata*) that are commonly found in Argentinean shelf waters (Almandoz et al., 2007) were not identified in the samples collected from PV.

*Pseudo-nitzschia* spp. were, in general, abundant in phytoplankton samples with densities ranging from 2200 cells $L^{-1}$ in Pardelas Beach, NG, to bloom values ranging from $1.6 \times 10^6$ cells $L^{-1}$ to $2.2 \times 10^6$ cells $L^{-1}$ in Bengoa and Larralde Beach, SJG, respectively, on November 11, 2010 (Table 1). These cell maximum densities were represented by *P. fraudulenta*, a species associated with DA production in PV area (Sastre et al., 2007), and to an unidentified species of the complex *P. pseudodelicatissima*. In spite of this, no cases of DA poisoning events resulting in wildlife mortality in PV have been documented to date.

Although various *Pseudo-nitzschia* species were found in the phytoplankton as well as in the fecal samples examined, co-occurrence of the same species in both kinds of samples was not always detected (Table 1). For example, while *P. fraudulenta* and an unidentified species of the complex *P. pseudodelicatissima* were the most abundant species in

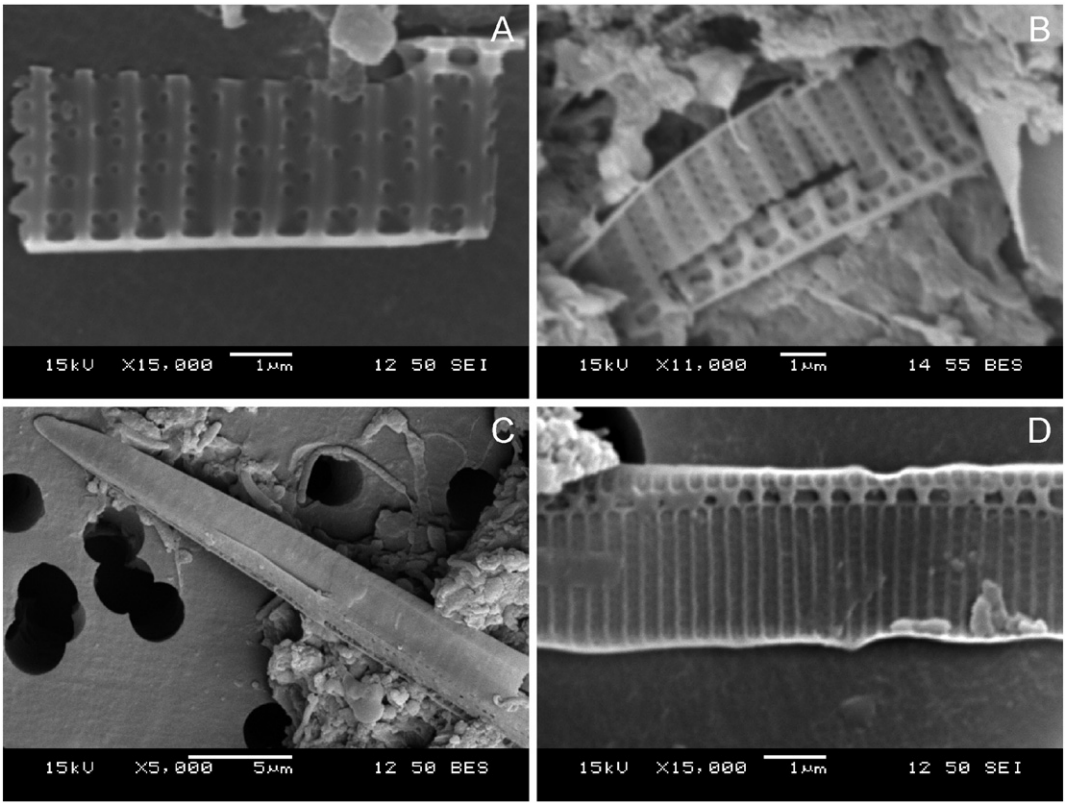


**Fig. 2.** Frustule fragments of *Pseudo-nitzschia* spp. observed in fecal samples of *Eubalaena australis* collected in their calving ground of Península Valdés. Scanning electron microscopy (SEM). *P. pungens* (A and B), *P. australis* (C) and the complex *P. pseudodelicatissima* (D).

the phytoplankton samples collected from SJG, *P. fraudulenta* was not found in any of the samples of stranded animals in SJG during the spring of 2010 (Table 1). Likewise, whereas *P. australis* was detected in fecal material from the stranded whale in Campamento 39 (SJG), it was not found in the phytoplankton samples collected two days later in the same area. This could be associated with differences in the phytoplankton sampling dates on account of the fact that the changes in phytoplankton composition are, in fact, fast and they can be detected from one week to another in PV area (Cadaillón, 2012).

Apart from diatom frustules, remains of copepods and other microcrustaceans were also found in all the fecal samples analyzed. Copepod remains were the most abundant in all whale feces with a clear dominance of copepodites' 5 (C5) mandibles of *Calanus australis* (D'Agostino et al., 2013), a common copepod in both NG and SJG. In line with this, previous research has revealed that *C. australis* is the most abundant copepod species along the southern coasts of Argentina and is extensively distributed in the inner and middle southern Patagonian shelf waters (Ramírez and Sabatini, 2000).

This study demonstrated for the first time the presence of potentially toxic *Pseudo-nitzschia* species in fecal samples from live and stranded individuals of *E. australis* that visited the calving ground of PV. This finding, together with the presence of microcrustacean remains (mainly C5 *C. australis* mandibles) observed in whale feces, indicates that *C. australis* may behave as vector of potentially toxic *Pseudo-nitzschia* spp. to whales when they feed on zooplankton patches in PV during spring. Copepods break diatom cells before their ingestion (Michels and Schnack-Schiel, 2005). Therefore, the presence of *Pseudo-nitzschia* frustule fragments instead of whole cells (as observed in phytoplankton samples) in all the fecal samples analyzed suggests that they derive from copepod gut contents.

Although our results are insufficient to assess if whales were exposed to DA or if toxin accumulation was the probable cause of death in stranded whales, it can be concluded that the presence of *Pseudo-nitzschia* spp. frustules in the fecal material analyzed as well as the occurrence of blooms of toxigenic species are both clear indicators that these animals could have been exposed to DA while they fed in the calving ground of PV.

Finally, it is also worthy of note that chronic exposure to DA gives rise to several types of diseases and pathologies in marine mammals (Brodie et al., 2006; Goldstein et al., 2009; Zabka et al., 2009). Thus, taking into account that exposure to phycotoxins is suggested as one of the hypotheses to explain mortality events among southern right whales in PV (Rowntree et al., 2013), and that it has also been reported to be one of the possible causes of reproductive failure in *E. glacialis* (Reeves et al., 2001; Durbin et al., 2002; Doucette et al., 2006; Leandro et al., 2010), further studies are clearly necessary to assess if southern right whales are indeed exposed to DA by means of toxin analyses of tissues, feces and plankton along time. These analyses will help assess the long-term exposure to this neurotoxin, which will, in turn, greatly increase our knowledge of the health of *E. australis* population in their calving ground.

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## References

Almandoz, G.O., Ferrario, M.E., Ferreyra, G.A., Schloss, I.R., Esteves, J.L., Paparazzo, F.E., 2007. The genus *Pseudo-nitzschia* (Bacillariophyceae) in continental shelf waters of Argentina (Southwestern Atlantic Ocean, 38–55°S). Harmful Algae 6, 93–103.

Brodie, E.C., Gulland, F.M.D., Greig, D.J., Hunter, M., Jaakola, J., St. Leger, J., Leighfield, T.A., Van Dolah, F.M., 2006. Domoic acid causes reproductive failure in California sea lions (*Zalophus californianus*). Mar. Mamm. Sci. 22, 700–707.

Cadaillón, A., 2012. Floraciones Algales Nocivas: Ficotoxinas en Fitoplancton y Zooplancton de los Golfos Nuevo y San José. Thesis Univ. Nac. Patagonia San Juan Bosco, Chubut. Argentina, p. 154.

Crespo, E.A., Pedraza, S.N., Dans, S.L., Coscarella, M.A., Svendsen, G.M., Degrati, M., 2014. Number of southern right whales *Eubalaena australis* and population trend in the neighbourhood of Península Valdés during the period 1999–2013 by means of aerial and boat surveys. Reporte IWC SRW Assessment Workshop SC/65b/BRG07.

D'Agostino, V.C., Hoffmeyer, M.S., Degrati, M., 2013. Análisis de fecas de individuos de *Eubalaena australis* en Península Valdés; *Calanus australis*: copépodo dominante en la dieta. Available in: http://www.sarem.org.ar/wp-content/uploads/2013/11/XXVI-JAM_Libro-de-Resumenes.pdf.

De La Riva, G.T., Johnson, C.K., Gulland, F.M., Langlois, G.W., Heyning, J.E., Rowles, T.K., Mazet, J.A., 2009. Association of an unusual marine mammal mortality event with *Pseudo-nitzschia* spp. blooms along the southern California coastline. J. Wildl. Dis. 45, 109–121.

Doucette, G.J., Cembella, A.D., Martin, J.L., Michaud, J., Cole, T.V.N., Rolland, R.M., 2006. Paralytic shellfish poisoning (PSP) toxins in North Atlantic right whales *Eubalaena glacialis* and their zooplankton prey in the Bay of Fundy, Canada. Mar. Ecol. Prog. Ser. 306, 303–313.

Durbin, E., Teegarden, G., Campbell, R., Cembella, A., Baumgartner, M.F., Mate, B.R., 2002. North Atlantic right whales, *Eubalaena glacialis*, exposed to paralytic sellfish poisoning (PSP) toxins via zooplankton vector, *Calanus finmarchicus*. Harmful Algae 1, 243–251.

Goldstein, T., Zabka, T.S., DeLong, R.L., Wheeler, E.A., Ylitalo, G., Bargu, S., Silver, M., Leighfield, T., Van Dolah, F., Langlois, G., Sidor, I., Lawrence Dunn, J., Gulland, F.M.D., 2009. The role of domoic acid in abortion and premature parturition of California sea lions (*Zalophus californianus*) on San Miguel Island, California. J. Wildl. Dis. 45, 91–108.

Gulland, F.M.D., Haulena, M., Fauquier, D., Langlois, G., Lander, M.E., Zabka, T., Duerr, R., 2002. Domoic acid toxicity in Californian sea lions (*Zalophus californianus*): clinical signs, treatment and survival. Vet. Rec. 150, 475–480.

Hasle, G.R., Fryxell, G.A., 1970. Diatoms: cleaning and mounting for light and electron microscopy. Trans. Am. Microsc. Soc. 89, 468–474.

Hoffmeyer, M.S., Lindner, M.S., Carribero, A., Fulco, V.K., Menéndez, M.C., Fernández Severini, M.D., Diodato, S.L., Berasategui, A.A., Biancalana, F., Berrier, E., 2010. Planktonic food and foraging of *Eubalaena australis*, on Península Valdés (Argentina) nursery ground. Rev. Biol. Mar. Oceanogr. 45, 131–139.

Leandro, L.F., Rolland, R.M., Roth, P.B., Lundholm, N., Wang, Z., Doucette, G.J., 2010. Exposure of the North Atlantic right whale *Eubalaena glacialis* to the marine algal biotoxin, domoic acid. Mar. Ecol. Prog. Ser. 398, 287–303.

Lefebvre, K.A., Powell, C.L., Busman, M., Doucette, G.J., Moeller, P.D.R., Silver, J.B., Miller, P.E., Hughes, S.S., Silver, M.W., Tjeerdema, R.S., 1999. Detection of domoic acid in northern anchovies and California sea lions associated with an unusual mortality event. Nat. Toxins 7, 85–92.

Lelong, A., Hégaret, H., Soudant, P., Bates, S.S., 2012. *Pseudo-nitzschia* (Bacillariophyceae) species, domoic acid and amnesic shellfish poisoning: revisiting previous paradigms. Phycologia 51, 168–216.

Lund, J.W.G., Kipling, C., LeGren, E.D., 1958. The inverted microscope method of estimating algal numbers and the statistical basis of estimations by counting. Hydrobiologia 11, 143–170.

Michels, J., Schnack-Schiel, S.B., 2005. Feeding in dominant Antarctic copepods — does themorphology of the mandibular gnathobases relate to diet? Mar. Biol. 146, 483–495.

Negri, R.M., Montoya, N.G., Carreto, J.I., Akselman, R., Inza, D., 2004. *Pseudo-nitzschia australis*, *Mytilus edulis*, *Engraulis anchoita*, and domoic acid in the Argentine Sea. In: Steidinger, K.A., Landsberg, J.H., Tomas, C.R., Vargo, G.A. (Eds.), Harmful Algae 2002. Florida Fish and Wildlife Conservation Commission, Florida Institute of Oceanography, and Intergovernmental Oceanographic Comission of UNESCO, St. Petersburg, Florida, USA, pp. 139–141.

Payne, R., 1995. Among Whales. Scribner, New York, NY, pp. 93–141.

Ramírez, F.C., Sabatini, M.E., 2000. The occurrence of Calanidae species in waters off Argentina. Hydrobiologia 439, 21–42.

Reeves, R.R., Rolland, R., Clapham, P.J., 2001. Causes of reproductive failure in North Atlantic right whales: new avenues of research. Report of a Workshop Held 26–28 April, 2000, Falmouth, MassachusettsNortheast Fisheries Science Center Reference Document 01–16. Woods Hole, MA, p. 46.

Rowntree, V.J., Uhart, M.M., Sironi, M., Chirife, A., Di Martino, M., La Sala, L., Musmeci, L., Mohamed, N., Andrejuk, J., McAloose, D., Sala, J.E., Carribero, A., Rally, H., Franco, M., Adler, F.R., Brownell Jr., R.L., Seger, J., Rowles, T., 2013. Unexplained recurring high mortality of southern right whale *Eubalaena australis* calves at Península Valdés, Argentina. Mar. Ecol. Prog. Ser. 493, 275–289.

Sastre, A.V., Santinelli, N.H., Esteves, J.L., Ferrario, M.E., 2001. Aspectos ecológicos de especies de *Pseudo-nitzschia* en aguas costeras patagónicas (Argentina). In: Alveal, K., Antezana, T. (Eds.), Sustentabilidad de la Biodiversidad. Universidad de Concepción, Chile, pp. 217–235.

Sastre, V., Santinelli, N., Marino, G., Solís, M., Pujato, L., Ferrario, M., 2007. First detection of domoic acid produced by *Pseudo-nitzschia* species, Chubut coastal waters, Patagonia, Argentina. Harmful Algae News 34, 12–14.

Sironi, M., 2004. Behaviour and Social Development of Juvenile Southern Right Whales (*Eubalaena australis*) and Interspecific Interactions at Península Valdés, Argentina. University of Wisconsin, Madison, p. 198 (PhD Dissertation).

Uhart, M., Rowntree, V.J., Sironi, M., Chirife, A., Mohamed, N., Pozzi, L., Franco, M., McAloose, D., 2009. Continuing southern right whale mortality events at Península Valdés, Argentina. Reporte IWC SRW Assessment Workshop SC/61/BRG18.

Zabka, T.S., Goldstein, T., Cross, R.W., Mueller, C., Kreuder Johnson, C., Gill, S., Gulland, F.M.D., 2009. Characterization of a degenerative cardiomyopathy associated with domoic acid toxicity in California sea lions (*Zalophus californianus*). Vet. Pathol. 46, 105–119.