



Microalgae at Two Pearl Oyster Farms in the Abrolhos Islands, Australia, and Implications for an Adductor Muscle Industry

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Abstract

The shellfish industry in Western Australia (WA) is expanding and, as part of the WA Shellfish Quality Assurance Program (WASQAP), microalgal populations need to be sampled. This is detailed in the WA Marine Biotoxin Monitoring and Management Plan (MBMMP) where alert levels for potentially toxic algae species have been set. The current research conducted water sampling over a 4-year period, from two pearl farms at the Abrolhos Islands, one near Rat Island and one near Pelsaert Island. Samples were Lugol preserved on-site and then analysed by Dalcon Environmental, Australia. Phytoplankton results included abundance data and percentage of the sample on all taxa, with mention of any potentially toxic microalgae. Only one sample revealed numbers of a potential toxin producer - (PTP, *Pseudo-nitzschia "seriata"* group H. Peragallo, 1900), above the alert level. Subsequent toxin analysis of frozen, flesh samples, taken on the same day, from cultured penguin pearl oysters, *Pteria penguin* (Röding, 1798), grown at the same site, were found to be negative for paralytic shellfish toxin (PST), amnesic shellfish toxin (AST), diarrhetic shellfish toxin (DST) and pectenotoxin-2 (PTX-2) by Symbio Labs in Sydney, NSW, Australia. Other phytoplankton sampling during this study, found very low levels of several other PTP's but with no incidences exceeding the MBMMP alert levels. The difficulty in identifying PTP's fully to species level and the variability in toxin production by closely related species poses significant challenges. Overall, there was some presence of potentially toxic microalgae in the Abrolhos Islands, with one incidence above alert levels. However, there was no evidence that biotoxins were present in the algae detected.

Keywords: phytoplankton, biotoxin, shellfish, sampling, monitoring

Introduction

Aquaculture is an expanding industry in Western Australia (WA) with shellfish culture becoming one of the fastest developing sectors in addition to seaweed farming. This growing interest in utilising coastal waters for aquaculture is also leading to a greater awareness of toxic algal species (Hallegraeff, 2004). UNESCO (2024) noted that every coastal country in the world is potentially affected by harmful phytoplankton blooms that can kill marine life and humans. Hallegraeff et al. (2021) noted that non-toxic species that attain high biomasses can also cause harmful algal blooms (HABs) by producing seawater discolorations, anoxia or mucilage. In northern WA, such an event

occurred in 1996 when silver-lip pearl oysters, *Pinctada maxima* (Jameson, 1901), suffered high mortalities in the presence of large blooms of *Trichodesmium erythraeum* Ehrenberg ex Gomont, 1892, an alga that was shown to be non-toxic but not a suitable food source (Negri et al., 2004).

As part of the requirements for shellfish aquaculture operations in WA, the monitoring of microalgae is required by WASQAP (WA Shellfish Quality Assurance Program) to analyse for the presence of potentially toxic microalgae. When consumed by shellfish, the toxins produced by these microalgae accumulate in the shellfish flesh and can be harmful to humans when the shellfish are consumed (Davies et al., 2016). Alert

levels have been established for the various potentially toxic micro-algal species, as detailed in the Marine Biotoxin Monitoring and Management Plan (MBMMP, WA DOH, 2016, 2020a). One of the problems encountered is that toxin producing species are similar in morphology and size to non-toxic ones (making species identification difficult), have similar physiological and ecological requirements and are randomly consumed by non-selective filter feeders. The subsequent accumulation of biotoxins is considered an unavoidable natural process and is determined by a range of factors including the relative abundance of toxin producers in a diverse plankton community and the residence time in the shellfish production area (EU Working Group, 2019). The ability to fully identify toxin producing species is therefore essential as is the knowledge of which closely related species produce toxins and those that do not. The difficulty in identifying different species (and strains) of potential toxin producers (PTP) under light microscopy was also highlighted in work by Verma et al. (2019) on *Prorocentrum* species. Some species produce okadaic acid (OA), dinophysistoxin (DTX) and derivatives which cause diarrhetic shellfish poisoning (DSP) while other very similar species do not produce any toxins. *Prorocentrum concavum* Y.Fukuyo, 1981 has been reported to contain OA previously (Dickey et al. 1990) but the strain sampled from NW Australia by Verma et al. (2019) was not found to contain any detectable amounts of OA or DTX and hence would not cause any DSP in shellfish. Similar issues occurred in the current study with the *Pseudo-nitzschia "seriata"* group.

As the subject of this report, it was hoped that a market for adductor muscles from penguin pearl oysters (*Pteria penguin*) (Röding, 1798) could be developed. Under the 2020 WASQAP Manual:

"This industry manual/user guide applies to all bivalve molluscan shellfish species commercially harvested or handled for the purpose of human consumption regardless of if they are harvested from the wild or from marine or land-based aquaculture facilities. Includes bivalve molluscan shellfish harvested for domestic and export markets. Bivalve molluscan shellfish species include, but are not limited to cockles, clams, mussels, oysters, pipis and unviscerated scallops, but do not include:

- Scallops and pearl oysters where the only part made available for human consumption is the adductor muscle and a hazard analysis approved by the DOH (Department of Health, WA) shows that heavy metals and/or marine biotoxins are not an identified hazard".

Given that our focus was on adductor muscles only, highly intensive sampling was not required by the DOH. This program collected microalgal water samples from two separate marine farm sites at the Abrolhos Islands (Fig. 1) over a 4-year period to examine the microalgal community and its variation over time. At the same

time, flesh was collected from cultured penguin pearl oysters and frozen in case alert levels of PTP's were detected and subsequent biotoxin analysis of the flesh was required. While harvesting of the whole soft tissues of shellfish would require much more intensive phytoplankton sampling in WA to accurately assess the toxin risk, this is not necessary for operations focused only on the shellfish adductor muscle.

Similarly, we did not need to determine toxin levels in different shellfish tissues but only in the portion to be consumed. The Food Standards Australia New Zealand (FSANZ, 2021) statement excludes scallops or pearl oysters where the only part consumed is the adductor muscle. They also state that toxin accumulation is lower in the adductor muscle. The statement notes that worldwide, there are a number of ongoing programs that monitor phytoplankton levels in harvesting waters and the levels of biotoxins in shellfish. Even so, recall events have still occurred with 26 recalls across all seafood in Australia between 2011 and 2020, and 15 notifications on the European Commission's bivalve molluscs portal referencing paralytic shellfish toxins (PST). It is important to note that various authors including Shumway and Cembella (1993), Shumway et al. (1995) and Bricelj and Shumway (1998) have found that phycotoxins are not distributed evenly throughout shellfish flesh but are usually concentrated in the mantle and digestive gland. Shumway and Cembella (1993) went on to state that scallop culture and commercial fisheries can still thrive in areas prone to toxic algal blooms if the adductor muscle is the only flesh utilised.

Existing knowledge and documented species of phytoplankton present in Western Australian waters is quite limited and there is a lack of baseline information upon which planning could be based. As a litre of seawater may contain up to a million microalgal cells, composed of at least 100–150 different species (Hallegraeff et al., 2010) it was important for this program to identify as fully as possible, the species present, their cell abundance and percentage of the total cells present. Rodriguez et al. (2024) noted that some species of microalgae cause shellfish poisoning syndromes such as amnesic shellfish poisoning (ASP), diarrhetic shellfish poisoning (DSP), paralytic shellfish poisoning (PSP) and neurotoxic shellfish poisoning (NSP), even at relatively low cell densities (e.g., 10^2 – 10^4 cells L^{-1}). Rodriguez et al. (2024) also noted that such low biomass harmful algal blooms (HAB's) can still cause lengthy harvesting bans for the shellfish industry due to their potential impact on consumers.

It is also important to note that a range of tropical invertebrate species from NW Australia, particularly crabs and molluscs, have been found to contain paralytic shellfish toxins (PST's) Negri and Llewellyn (1998). Paralytic shellfish toxins were found in 18 species of crabs and molluscs including 8 species that had not previously been recorded as toxic. These animals were considered to act as biofilters that

magnified naturally occurring, but unidentified PST's in the region (Negri and Llewellyn, 1998).

In the current study particular attention was paid to potentially toxic species and these were identified as fully as possible. Heavy metal sampling was not considered in this paper. The presence of the potentially toxic diatom *Pseudo-nitzschia* was detected but it could only be identified to the "*seriata*" group using a light microscope, as all species have similar morphologies (Rhodes, 1998). The *Pseudo-nitzschia* "*seriata*" group includes some 20 species which share the most prominent ecological and morphological features of the type species *Pseudo-nitzschia seriata* (Hallegraeff et al., 1995). Several *Pseudo-nitzschia* species belonging to the "*seriata*" group have been implicated in various amnesic shellfish poisonings (ASP) since 1987 (Hallegraeff et al., 1995). The toxin produced by microalgae (diatoms) of the genus *Pseudo-nitzschia* is the neurotoxin domoic acid.

Sampling plans that monitor potential toxin producer's (PTP's) in shellfish production areas should therefore be capable of assessing the spatial and temporal variation of the risks of toxins occurring in molluscs. This is a difficult task given the regional and seasonal complexities driving phytoplankton communities in any area. Nevertheless, the sampling plans need to allow analyses that are representative of the areas of interest (EU Working Group, 2019). This is the dilemma faced by phytoplankton monitoring at the Abrolhos Islands that can allow harvesting of shellfish for human consumption. Hence, a balance between a logical cost-effective sampling programme and the risk to human health was made. In the current research, a combination of extended, ad-hoc phytoplankton sampling was used in association with mollusc flesh samples to assess the presence and abundance of PTP's and also whether any toxins were actually produced when PTP's were present in significant numbers.

Materials and Methods

Ethical approval

The study was conducted in accordance with the WA Shellfish Quality Assurance Program (WASQAP) Operations Manual, 6th edition (WA DOH, 2017), the WA Department of Health Marine Biotoxin and Monitoring Plan, Version 1 (2016), and the Australian Code for the Care and Use of Animals for Scientific Purposes, 8th edition (2013).

Sampling site

Located 60–80 km off the coast of Geraldton, WA, are the Abrolhos Islands where the marine farms are situated (Fig. 1). Latitude Pearls established Licence 1540 in the Pelsaert Group of Islands in 1998 with Licence 1574 established at a similar time in the Easter Group (Figures 1 and 2). The existing shellfish farm licence 1574 was taken over by Abrolhos Aquaculture

Australia (AAA) in 2016. The companies already produce Akoya and Black-lipped pearls in the pristine Abrolhos Islands waters using some of the lowest impact (longline) aquaculture techniques in the world. Latitude Pearls and AAA are currently permitted to culture the bat wing or penguin pearl oyster (*Pteria penguin*) and a number of other species not mentioned herein. *Pteria penguin* spat have previously been brought onto each farm from the AAA hatchery at Rat Island and these have been under cultivation during the period, 2019–2023. Other settled spat and juveniles of various species are also naturally present in the culture gear and on longlines.

The Latitude Pearls site is located on the west side of Pelsaert Island in the Southern Group of the Abrolhos Islands, some 10 km from the occasionally inhabited islands of the Group (Figures 1 and 2). The depth of the site ranges from approximately 6 m to 15 m depth, and the underlying seabed is primarily sand. Water movement in the area is significant with current direction at this site heavily determined by the tidal flows. The AAA sites (Hatchery and jetty Facilities and Lease 1574) are located on and near Rat Island, Easter Group, at the Abrolhos Islands (Figures 1 and 2). The 1574 marine farm site is located at least 1.5 nm or 2,800 m from any habitable facilities on Rat Island or other nearby islands. Commercial fishermen and aquaculture proponents occasionally occupy shacks on Rat Island and some nearby islands. The farm site is up to 40 m deep with a sandy bottom with some coral outcrops located throughout the farm. Much of the site is 10–15 m deep but does have some shallower areas of only several metres deep. Water movement in the area is significant with the habitable areas having no detectable impact on the farm site.

Environmental monitoring of some basic parameters has been carried out for many years under the Department of Primary Industries and Regional Development (DPIRD) required Management and Environmental Monitoring Plan (MEMP) for each farm. After various sampling and testing was conducted over 4 years, it was anticipated that the farm sites would be successfully assessed under the DOH requirements such that adductor muscles from the penguin oyster (*Pteria penguin*) could be sold for food (human consumption). The adductor muscles resemble those from the saucer scallop that is caught nearby as the basis of a wild fisheries resource. That species grows in surrounding waters and has been marketed and eaten by humans for many years with no apparent health concerns related to algae.

As noted earlier, the current study was focused on satisfying DOH requirements for the safe production, harvesting and sale of adductor muscles from the penguin pearl oyster (*Pteria penguin*).

The Abrolhos Islands consist of 122 islands and reefs on the edge of the continental shelf some 60 km offshore from Geraldton, WA (DOF, 2012a). They extend



Fig. 1. Houtman Abrolhos Islands overview map (DPIRD, 2022).

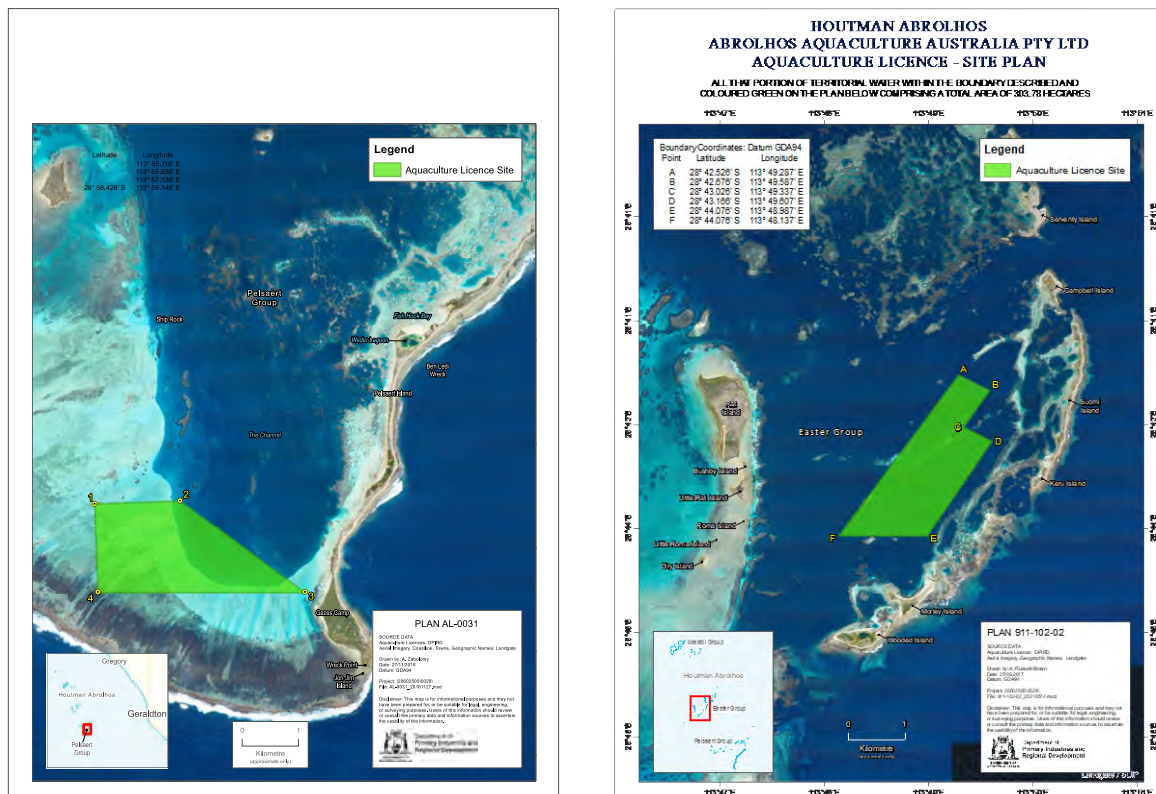


Fig. 2. Location of Pelsaert (WA) Pty Ltd site (1540) and Abrolhos Aquaculture Australia Pty Ltd, Rat Island site (1574) at the Abrolhos Islands.

for more than 100 km between latitudes 28°15'S and 29°00'S and are comprised of three major island groups separated by channels of around 40 m deep: Wallabi-North Island Group; Easter Group and Pelsaert (or Southern) Group (DOF, 2012a). DOF (2012b) states that the Abrolhos Islands attracts significant social and economic activity including commercial fisheries for rock lobster, scallops and finfish as well as pearling aquaculture and scientific research.

The islands lay in the path of the southward flowing Leeuwin Current. The current funnels warm tropical water from the Pacific Ocean, southwards past Indonesia and down along Western Australia's continental shelf. This is the only poleward flowing current of any eastern ocean boundary globally, all other eastern boundary currents flow towards the equator. Hence, the unique Leeuwin Current means relatively warm water temperatures year-round, allowing a range of tropical species to exist at unusually high latitudes (Collins et al., 1991). Whilst winter water temperatures are usually around 18–20 °C, this rises to 22–23 °C in autumn or spring and may exceed 28 °C in summer at which times the pearl shells grow quickly. The Leeuwin Current flows year-round but its flow is strongest in autumn and winter (Rousseaux, 2011, 2018) with nutrients in the surface waters off Ningaloo Reef (some 200 km north of the Abrolhos Islands) found to increase at that time of year, thereby supporting more phytoplankton production. A combination of an accelerating Leeuwin Current and net sea surface cooling was shown to increase the mixed layer depth resulting in increased nutrient and chlorophyll *a* concentration in the photic zone and the subsequent increase in phytoplankton productivity (Rousseaux, 2011).

An overview of environmental conditions at the Abrolhos Islands, relevant to Latitude Pearls and AAA, is found in Fisheries Research and Development Corporation (FRDC) Project 2007/216 – Develop the Non-Maxima Pearl Industry at the Abrolhos Islands (Cropp et al., 2011).

It should be noted that previous culture operations and extensive environmental monitoring (mentioned above) have shown the established Abrolhos Island aquaculture sites to be suitable for pearl oyster culture activities, including the penguin oyster *Pteria penguin*. There is no supplementary feeding of pearl oysters, all feed is naturally occurring in the form of phytoplankton. Various research has shown that small scale or relatively low-medium density shellfish farming is relatively benign and has no detectable impact on the environment.

Environmental parameters

For the current program the monitoring of relevant environmental parameters by Latitude/Pelsaert (WA) and AAA Pty Ltd was limited to water temperature at various depths using a hand-held, portable instrument.

As part of the WA MEMP requirements to identify, monitor and minimise potential environmental impacts from pearl oyster and other marine farming activities, temperature data was manually recorded from two locations on each aquaculture farm site/licence. Additionally, as mentioned, a third site outside the farm site and not likely to be affected by farming activities was used as a reference site for comparison. This site was monitored at the same time as the farm sites. For the current study, only relevant water temperature data from Site 2 at each farm site was utilized to provide spot data for the phytoplankton sampling. Accurate GPS positions were noted for each sampling location, and these same positions were monitored on each subsequent monitoring trip.

Other parameters such as pH, dissolved oxygen and sediment pH were separately monitored as part of the routine MEMP but were not reported herein. Overall, the documentation of phytoplankton presence and abundance data is expected to be highly beneficial to the developing shellfish industry in WA and potentially Australia as a whole, given the present lack of published phytoplankton information.

A total of nine 1 L water samples were collected from Site 2 at Marine Farm 1540 near Pelsaert Island at the Abrolhos Islands over the period 18 May 2019 to 04 November 2023. Specifically, sample dates were 18 May 2019, 11 September 2019, 21 May 2020, 06 September 2020, 16 March 2021, 11 June 2021, 17 March 2022, 08 July 2022 and 04 November 2023.

A total of nine 1 L water samples were collected from Site 2 at Marine Farm 1574 near Rat Island at the Abrolhos Islands over the period 18 May 2019 to 03 November 2023. Specifically, sample dates were 18 May 2019, 11 September 2019, 21 May 2020, 08 November 2020, 22 March 2021, 11 June 2021, 16 March 2022, 07 July 2022 and 03 November 2023. The water samples were collected at the same site/farm area being used for cultivation of the penguin pearl oyster (*Pteria penguin*).

The microalgae or phytoplankton water samples were collected using the integrated tube method of >2m depth, as detailed by the WASQAP and MBMMP manual and Dalcon Environmental (WA DOH, 2020a, b; Dalcon Environmental, 2019). All samples were immediately preserved with Lugol's iodine solution after collection and labelling and transported to the Dalcon Environmental Lab as per MBMMP guidelines.

Upon arrival at the lab and, after sample integrity was assessed, the sample was prepared for analysis. The sample was concentrated from 1,000 mL to 10 mL (100× concentration) using gravity assisted filtration. The sample was then analysed microscopically using Dalcon Environmental in-house method DE-AM-01 (Dalcon Environmental, 2019) at a total magnification of between 100× and 400×. All microalgal taxa were identified and enumerated with a limit of reporting between 10 cells L⁻¹–200 cells L⁻¹. Alert levels for the

various microalgal species, as detailed in the MBMMP (WA DOH, 2020a), are shown in Table 1. This table summarises the phytoplankton levels (in cells L⁻¹) that trigger management action.

It should be noted that several amendments should be considered for Table 1. In revised taxonomy work by John et al. (2014), the PTP *Alexandrium tamarens* was found to be non-toxic. Additionally, *Alexandrium catenella* is a coldwater species only found in Tasmania and hence should probably be deleted from the Table. The species *Alexandrium pacificum* however, should be included on this Table as it has been the major PSP species occurring in New South Wales (NSW) (Barua et al., 2020) and the Perth region

(Trayler and Cosgrove, 2024). These issues need to be examined carefully as there are both monitoring and health/food safety implications for WA.

If phytoplankton sampling detected an exceedance of alert levels noted above, then flesh testing for biotoxins in shellfish cultured in these waters would become necessary. Hence, to pre-empt any potential requirement for this, flesh of around 200 g wet weight in total, was collected from 20 individual, cultured *Pteria penguin* adults from each site and frozen.

The threshold limits for the various marine toxins, extracted from the WASQAP (WA DOH, 2020b) manual are included in Table 2.

Table 1. Phytoplankton alert levels for marine biotoxins (most relevant), summarising the cell concentrations (cells L⁻¹) that trigger management action (MBMMP, WA DOH, 2020a).

Microalgae species	Type of toxin	Alert level	Alert level to initiate flesh testing (cells L ⁻¹)
<i>Alexandrium catenella</i> ¹	PSP	100	200
<i>Alexandrium minutum</i> ¹	PSP	100	200
<i>Alexandrium ostenfeldii</i> ¹	PSP	100	200
<i>Alexandrium tamarens</i> ¹	PSP	100	200
<i>Gymnodinium catenatum</i>	PSP	500	1,000 (mussels) 2,000 (other shellfish)
<i>Dinophysis acuminata</i>	DSP	1,000	1,000
<i>Dinophysis acuta</i>	DSP	500	1,000
<i>Dinophysis caudata</i>	DSP	500	1,000
<i>Dinophysis fortii</i>	DSP	500	1,000
<i>Prorocentrum lima</i>	DSP	500	500
<i>Pseudo-nitzschia</i> "seriata" group (<i>P. multiseriata</i> and <i>P. australis</i>) ²	ASP	50,000*	500,000
<i>Pseudo-nitzschia</i> "delicatissima" group ²	ASP	500,000	500,000
<i>Karenia brevis</i>	NSP	500	1,000

*At 50,000, weekly flesh testing will commence. N.B. The cell levels within each toxin group are cumulative.

¹*Alexandrium* species may be difficult to identify when numbers are low. If any doubt exists, they should be treated as potentially toxic.

²Species within the *Pseudo-nitzschia* groups are difficult to identify. The group includes *P. australis*, *P. pungens* (Grunow ex Cleve) Hasle, 1993 and *P. multiseriata*. The *Pseudo-nitzschia* "delicatissima" group includes *P. turgidula* (Hustedt) Hasle, 1993, *P. fraudulenta* (Cleve) Hasle, 1993, *P. delicatissima* (Cleve) Heiden, 1928, *P. delicatissima* (Hasle) Hasle, 1993 and *P. multistriata* (H.Takano) H.Takano, 1995.

Table 2. Biotoxin threshold limits WASQAP (WA DOH, 2020b).

Toxin type (Equivalent compound)	Sampling frequency	Threshold limit	Regulatory standard
PSP toxins (saxitoxins equivalent)	monthly	0.8 mg kg ⁻¹	FSANZ Std 1.4.1 c
ASP toxins (domoic acid equivalent)	monthly	20 mg kg ⁻¹	FSANZ Std 1.4.1 c
DSP toxins (okadaic acid equivalent)	monthly	0.2 mg kg ⁻¹	FSANZ Std 1.4.1 c
NSP toxins	monthly	200 MU kg ⁻¹	FSANZ Std 1.4.1 c

FSANZ: Food Standards Australia New Zealand.

Results

The complete results from our phytoplankton samples, as analysed by Dalcon Environmental, are shown in Table 3 (Pelsaert Island Licence 1540) and Table 4 (Rat Island-Licence 1574). In brief, there was only one exceedance of alert or trigger values found in any samples from 18 May 2019 through to 4 November 2023 and this was from Rat Island on 18 May 2019.

Analysis of the phytoplankton sample taken on 18 May 2019 from the Easter Group (near Rat Island) showed that cell density of *Pseudo-nitzschia "seriata"* group was in exceedance of the alert level to initiate flesh testing of 50,000 cells L⁻¹ (on a weekly basis) as per the Marine Biotoxin Monitoring and Management Plan (MBMMP, WA DOH, 2016). Subsequently, as required under the MBMMP, whole flesh samples from penguin oysters grown at site 1574 and previously frozen, were sent to Symbio Laboratories in Sydney (NSW) for biotoxin analysis. These results are shown in Table 5 and were well below toxin threshold limits.

While there were no other PTP's detected above alert levels, there was a regular presence of both *Pseudo-nitzschia "seriata"* group and *Pseudo-nitzschia "delicatissima"* group at both marine farm sites (Tables 3 and 4) but with maximum densities for the other sample dates of around 36,000 cells L⁻¹ and 70,000 cells L⁻¹, respectively. These maximum densities both occurred at Pelsaert Island in March of 2022, with a water temperature of around 26 °C (Table 6). For other PTP's, there were no other significant detections above 800 cells L⁻¹, at either site in the 9 samples collected.

It is interesting to note that in May 2019, with water temperatures of around 20 °C, there was an apparent bloom of *Oscillatoria* spp. (now known as *Trichodesmium*) at Pelsaert Island. Density was around 217,000 cells L⁻¹ with an abundance of 71 % of the phytoplankton sample, whilst further north at the deeper Rat Island site, the density was only 62,300 cells L⁻¹ with an abundance of only 25 % in the sample. No apparent health issues were noted in the penguin oysters being cultured at either site. There were no other occurrences of *Oscillatoria* spp. detected in any samples except for a minor presence (500 cells L⁻¹) in November 2023 at Rat Island when the water temperature was around 23 °C (Table 7).

Discussion

While our results did not show any toxicity from the *Pseudo-nitzschia "seriata"* group present in this case, it is important to identify as fully as possible, any *Pseudo-nitzschia* present. Several species produce high concentrations of domoic acid (ASP toxins) per cell while others are non-toxic (Rhodes, 1998), as in this sample. The clear need to fully identify all PTP's to species level, and the associated need to assess the toxin producing capability of each species is again

apparent. Phytoplankton monitoring work in the EU has also noted the difficulties in accurately assessing the biotoxin risks in shellfish production areas, given the highly variable nature of the phytoplankton communities, although the importance of conducting effective sampling programs remains essential (EU Working Group, 2019).

In our monitoring program, there were no other exceedances of alert levels in phytoplankton samples, hence, there was no need to send further samples for biotoxin analysis. Over the 4-year microalgal sampling program, low to very low levels of several other PTP's were found within some of the samples from each site. These included *Pseudo-nitzschia "delicatissima"* group H. Peragallo, 1900, *Alexandrium* spp., and *Dinophysis acuminata*. Even so, none of these PTP's occurred at both sites in all samples and were more commonly found at very low levels if at all. Table 8 lists some of the known toxins and their associated algal species, including those related to or similar to the taxa observed at the Abrolhos Islands during our sampling.

In recent years we have seen the establishment of the Harmful Algal Information System (HAIS; <https://hab.ioc-unesco.org/>) in collaboration with the International Oceanographic Data and Information Exchange (IODE) of the Intergovernmental Oceanographic Commission (IOC) of UNESCO. This is in response to the need for an authoritative and co-ordinated worldwide online source of information about harmful algae (Hallegraeff et al., 2021). The HAIS consists of two main databases that document the geographical range of harmful species and the database recording harmful algal events; both are continuously revised and updated (Hallegraeff et al., 2021). Such databases are essential to improving our knowledge of particular microalgal species and their potential toxicity, wherever they may occur.

Although only a relatively small dataset, the current results can provide valuable additions to large scale spatial and temporal studies (Davies et al., 2016) via collation into the Australian Phytoplankton Database. This database is available through the Australian Ocean Data Network (AODN: <https://portal.aodn.org.au/>). It is the main repository for all marine data in Australia and is updated with new data that automatically uploads to the AODN portal (Davies et al., 2016).

Hallegraeff and Jeffrey (1994) found that the phytoplankton on shelf waters of the Gulf of Carpentaria, Arafura Sea, Timor Sea and NW Australia is basically a diatom flora and distinctly different from the predominantly dinoflagellate flora of the Coral Sea and Indian Ocean. Their diatom flora exhibited species similarly recorded in the current study such as *Bacteriastrum* spp., *Chaetoceros* spp., *Coscinodiscus* spp., *Nitzschia* spp., *Rhizosolenia* spp. and *Thalassiosira* spp. Diatoms (Bacillariophyceae) also

Table 3. Summary of Pelsaert Island phytoplankton data (cells L⁻¹).

REPORT NUMBER	DE00947.1		DE01045.1		DE01383.1		DE01505.1		DE01796.1		DE01813.1		DE02150.2		DE02288.2		DE02906.2	
SITE	Pelseart Group		Pelseart Group		Pelseart Group		Pelseart Group		Pelseart Group		Pelseart Group		Pelseart Group		Pelseart Group		Pelseart Group	
DATE SAMPLED	18-05-19		11-09-19		21-05-20		06-11-20		16-03-21		11-06-21		17-03-22		07-07-22		04-11-23	
DATE RECEIVED	24-05-19		19-09-19		05-06-20		11-11-20		04-06-21		23-06-21		30-03-22		20-07-22		13-11-23	
DATE ANALYSED	01-06-19		17-10-19		10-07-20		18-11-20		08-06-21		23-06-21		07-04-22		27-07-22		14-11-23	
Bacillariophyceae	59000	19.41	165900	99.22	56900	98.78	10100	99.02	14000	100.00	7800	97.50	270800	97.69	14400	39.78	144700	74.47
Achnanthes spp.	300	0.10					300	2.94										
Achnanthidium spp.																		
Amphipleura spp.											100	1.25						
Amphora spp.	2000	0.66	1900	1.14	100	0.17	600	5.88			200	2.50	800	0.29	1400	3.87		
Asterionellopsis glacialis			7700	4.61														
Bacillaria spp.																	100	0.05
Bacteriastrium spp.			1900	1.14							400	5.00	1200	0.43	400	1.10		
Bleakeleya spp.	300	0.10			500	0.87	100	0.98	200	1.43	100	1.25						
Cerataulina pelagica					300	0.52												
Cerataulina spp.											200	2.50						
Chaetoceros spp.			65700	39.29	400	0.69					1000	12.50	156000	56.28	2000	5.52	45400	23.37
Climacosphenia spp.															800	2.21		
Cocconeis spp.	25000	8.22	2100	1.26	100	0.17	500	4.90	100	0.71	1000	12.50			200	0.55	100	0.05
Coscinodiscus spp.											100	1.25						
Cyclotella spp.					500	0.87												
Cylindrotheca closterium	2000	0.66	5300	3.17	3100	5.38	500	4.90					800	0.29	400	1.10	26000	13.38
Cylindrotheca spp.									2500	17.86	200	2.50						
Cymbella spp.							200	1.96										
Dactylosolen spp.																	300	0.15
Ditylum spp.															200	0.55		
Diploneis spp.	100	0.03	1100	0.66							200	2.50						
Entomoneis spp.																	100	0.05
Eucampia spp.			100	0.06	800	1.39							800	0.29			100	0.05
Fallacia spp.							200	1.96										
Fragilaria spp.							200	1.96			300	3.75						
Fragilariopsis spp.	2000	0.66																
Guinardia spp.																	300	0.15
Guinardia striata			1500	0.90														
Hantzschia spp.	1000	0.33	100	0.06			200	1.96										
Hemialus spp.	400	0.13	200	0.12														
Leptocylindrus spp.																	200	0.10
Leptocylindrus danicus			17900	10.71									2000	0.72	400	1.10		
Licmophora lyngbyei			1900	1.14														
Licmophora spp.	1000	0.33					1000	9.80	400	2.86	200	2.50					400	0.21
Lioloma spp.					200	0.35												
Lithodesmium spp.			100	0.06														
Luticola spp.							100	0.98										
Mastoglaia spp.	1000	0.33					100	0.98	400	2.86								
Melosira spp.																		
Microtabella spp.					100	0.17					100	1.25			200	0.55		
Navicula transitrans var. derasa																		
Navicula spp.	17000	5.59			300	0.52	1700	16.67	1300	9.29	900	11.25			1200	3.31	3100	1.60
Nitzschia longissima	3000	0.99	7100	4.25	100	0.17			1200	8.57			1200	0.43	2000	5.52	34000	17.50
Nitzschia spp.	1000	0.33			2800	4.86	4200	41.18	7500	53.57	2800	35.00			200	0.55		
Pinnularia spp.									200	1.43								
Plagiodiscus spp.							200	1.96										
Plagiogrammopsis spp.	300	0.10																
Plagiotropis spp.																		
Pleurosigma spp.	1000	0.33							100	0.71					200	0.55		
Pseudo-nitzschia "delicatissima" group (PTP)			31300	18.72	42700	74.13							69600	25.11	1400	3.87	8200	4.22
Pseudo-nitzschia "seriata" group (PTP)			100	0.06									36000	12.99	400	1.10	3600	1.85
Rhizosolenia setigera					700	1.22												
Rhizosolenia spp.	500	0.16	1700	1.02														
Skeletonema spp.			9800	5.86														
Surirella spp.	100	0.03													2000	5.52	20400	10.50
Synedra spp.									100	0.71								
Thalassionema spp.					4200	7.29							1200	0.43	200	0.55	2000	1.03
Thalassiosira pseudonana															600	1.66		
Thalassiosira spp.			1500	0.90									800	0.29			300	0.15
Thalassiothrix spp.	1000	0.33	6800	4.07									400	0.14			100	0.05
Tryblionella spp.			100	0.06														
Chlorodendrophyceae															13000	35.91	15000	7.72
Tetraselmis spp.															13000	35.91	15000	7.72
Cryptophyceae															2000	5.52	3700	1.90
Cryptomonas spp.															200	0.55		
Hillea spp.															1800	4.97	3300	1.70
Rhodomonas spp.																	400	0.21
Chrysophyceae																	5000	2.57
Chrysoschromulina spp.																	5000	2.57
Pachysphaera spp.																		
Cyanobacteria	245000	80.59													1000	2.76		
Oscillatoria spp.			217000	71.38														
Pseudonabaena spp.	28000	9.21													1000	2.76		
Dictyochophyceae			100	0.06														
Octatis octonaria			100	0.06														
Dinophyceae			1200	0.72	700	1.22	100	0.98			100	1.25	2800	1.01	4600	12.71	5800	2.99
Akashiwo sanguinea			100	0.06														
Ceratium lineatum			100	0.06														
Ebria tripartita			100	0.06														
Gonyaulax spp.													400	0.14				
Gymnodinium spp.			200	0.12													4500	2.32
Gyrodinium spirale											100	1.25						
Gyrodinium spp.			100	0.06											3200	8.84	200	0.10
Heterocapsa spp.					300	0.52	100	0.98							400	1.10	300	0.15
Karenia/Karlodinium/Gymnodinium group (PT)													800	0.29	800	2.21		
Karlodinium spp.					100	0.17												
Oxytoxum spp.																	100	0.05
Peridinioid Dinoflagellate (unknown)															200	0.55		
Prorocentrum dentatum			100	0.06	200	0.35												
Prorocentrum micans			100	0.06														

Table 4. Summary of Rat Island phytoplankton data (cells L⁻¹).

REPORT NUMBER	DE00947.2		DE01045.2		DE01383.2		DE01505.2		DE01796.2		DE01813.2		DE02150.1		DE02288.1		DE02906.1	
SITE	Rat Island		Rat Island		Rat Island		Rat Island		Rat Island		Rat Island		Rat Island		Rat Island		Rat Island	
DATE SAMPLED	18-05-19		11-09-19		21-05-20		08-11-20		22-03-21		11-06-21		16-03-22		07-07-22		03-11-23	
DATE RECEIVED	24-05-19		19-09-19		05-06-20		11-11-20		04-06-21		23-06-21		30-03-22		20-07-22		13-11-23	
DATE ANALYSED	01-06-19		17-10-19		10-07-20		18-11-20		08-06-21		23-06-21		07-04-22		27-07-22		14-11-23	
Bacillariophyceae	181100	73.32	72000	99.04	21800	94.37	121400	99.51	4500	75.00	8500	97.70	406800	63.72	104600	90.45	47800	60.43
Achnanthes spp.			100	0.14													100	0.13
Achnanthisium spp.							200	0.16										
Amphora spp.	1900	0.77	700	0.96			200	0.16			100	1.15	400	0.06			6000	7.59
Asterionellopsis glacialis			200	0.28											400	0.35		
Asterionellopsis spp.											200	2.30						
Bacillaria spp.			200	0.28														
Bacteriastrium spp.	1100	0.45	2500	3.44	300	1.30	400	0.33			2000	22.99						
Bleakeleya spp.	200	0.08	100	0.14	200	0.87												
Campylodiscus spp.							100	0.08										
Campylosira spp.							400	0.33										
Cerataulina pelagica					600	2.60												
Chaetoceros spp.	32300	13.08	9300	12.79	3900	16.88	39500	32.38			2800	32.18	33600	5.26	34600	29.92	1500	1.90
Cocconeis spp.	5900	2.39	11800	16.23			100	0.08					400	0.06			400	0.51
Coscinodiscus spp.			200	0.28			100	0.08			100	1.15			200	0.17		
Cyclotella spp.					100	0.43												
Cylindrotheca closterium	12100	4.90	2700	3.71	400	1.73	1100	0.90					8000	1.25	10000	8.65	8400	10.62
Cylindrotheca spp.									300	5.00	500	5.75						
Dactylosolen spp.							100	0.08							200	0.17	400	0.51
Ditylum spp.							100	0.08										
Diploneis spp.			100	0.14														
Entomoneis tenuistriata			100	0.14														
Eucampia spp.					200	0.87	700	0.57							2000	1.73		
Fragilaria spp.															3800	3.29		
Grammatophora spp.																	2100	2.65
Guinardia spp.																	500	0.63
Guinardia striata	900	0.36																
Gyrosigma spp.							100	0.08										
Hantzschia spp.			200	0.28														
Hemialus spp.															1000	0.86		
Leptocylindrus spp.																	600	0.76
Leptocylindrus danicus	9700	3.93	200	0.28											400	0.35		
Licmophora lyngbyei			500	0.69														
Licmophora spp.	800	0.32					200	0.16	200	3.33							9700	12.26
Lithodesmium spp.			100	0.14														
Mastogloia spp.			100	0.14			100	0.08	200	3.33								
Melosira spp.																		
Meuniera membranacea															400	0.35		
Navicula transitrans var. derasa			100	0.14											800	0.69		
Navicula spp.	5900	2.39			200	0.87	100	0.08	600	10.00	200	2.30	800	0.13	4200	3.63	7300	9.23
Nitzschia longissima	3700	1.50	400	0.55					100	1.67			44000	6.89	5400	4.67	8300	10.49
Nitzschia spp.	100	0.04	300	0.41	1500	6.49	59000	48.36	1600	26.67	1100	12.64						
Oestrupia spp.							200	0.16										
Pinnularia spp.							100	0.08										
Plagiodiscus spp.			300	0.41			300	0.25	100	1.67								
Pleurosigma spp.	7700	3.12	100	0.14					100	1.67								
Pseudo-nitzschia "delicatissima" group (f)	12100	4.90	1100	1.51	12800	55.41	13900	11.39			200	2.30	59200	9.27	22600	19.54	400	0.51
Pseudo-nitzschia "seriata" group (PTP)	84300	34.13	9100	12.52					300	5.00	100	1.15	26000	4.07	5200	4.50		
Rhizosolenia setigera											100	1.15						
Rhizosolenia spp.	1100	0.45	200	0.28	300	1.30	500	0.41							1200	1.04		
Rhopalodia gibba									800	13.33	100	1.15						
Skeletonema costatum											800	9.20						
Skeletonema spp.			30700	42.23			2800	2.30									1400	1.77
Striatella spp.							100	0.08										
Synedra spp.									200	3.33								
Thalassionema spp.	500	0.20	200	0.28	1300	5.63	1000	0.82			200	2.30	2000	0.31	8600	7.44	200	0.25
Thalassiosiphysa spp.			100	0.14														
Thalassiosira pseudonana	100	0.04													1800	1.56	100	0.13
Thalassiosira spp.			100	0.14									232000	36.34			300	0.38
Thalassiothrix spp.	700	0.28	100	0.14											1000	0.86	100	0.13
Toxarium spp.															600	0.52		
Tryblionella spp.			100	0.14														
Chlorodendrophyceae																	6900	8.72
Tetraselmis spp.																	6900	8.72
Chlorophyceae					800	3.46												
Pediastrum spp.					800	3.46												
Cryptophyceae													92000	14.41			1800	2.28
Cryptomonas spp.													92000	14.41			1600	2.02
Hillea spp.																	200	0.25
Rhodomonas spp.																		
Chrysophyceae																	500	0.63
Chrysochromulina spp.																	400	0.51
Pachysphaera spp.																	100	0.13
Cyanobacteria	62300	25.22															2000	2.53
Oscillatoria spp. (PT) (t)	62300	25.22															500	0.63
Pseudanabaena spp.																	1400	1.77
Trichodesmium spp. (t)																	100	0.13
Dictyochophyceae			200	0.28	200	0.87	100	0.08			100	1.15	3600	0.56	400	0.35		
Dictyocha fibula var. rhombica															200	0.17		
Dictyocha spp.											100	1.15						
Octatis octonaria			200	0.28	200	0.87	100	0.08					3600	0.56	200	0.17		
Dinophyceae	3500	1.42	500	0.69	300	1.30	400	0.33	1500	25.00	100	1.15	8060	1.26	10640	9.20	1300	1.64
Alexandrium spp. (PTP)			100	0.14														
Ceratium furca									200	3.33								
Ceratium trichoceros									100	1.67								
Dinophysis acuminata (PT)									200	3.33								
Gonyaulax spp.													60	0.01	40	0.03		
Gymnodinium spp.													1200	0.19				
Gyrodinium spirale																		

Table 5. Biotoxin results from penguin oyster whole flesh samples, 18 May 2019 (Shellfish, Method SE6038-OP, Symbio Labs, 2019).

Lab ID	Date received	Sample date	Test analysis	Results	Units
S808775-2	24/7/2019	18/5/2019	Total PSP screen	<0.025	mg STX eq.kg ⁻¹
S808775-2	24/7/2019	18/5/2019	Domoic acid	<1.0	mg kg ⁻¹
S808775-2	24/7/2019	18/5/2019	Total DSP	<0.015	mg OA eq.kg ⁻¹
S808775-2	24/7/2019	18/5/2019	Pectenotoxin-2	<0.025	mg PTX2 eq.kg ⁻¹

Table 6. Pelsaert Island Marine Farm 1540, Site #2 water temperatures (°C). Latitude Farm Site #2: Lat. 28°57.720'S, Long. 113°56.015'E.

Date	Surface	Mid	Bottom
18/05/2019	20.6	20.1	19.4
21/05/2020	21.9	20.3	20.3
31/07/2020	22.4	21.5	21.5
6/11/2020	21.3	21.2	21.5
16/03/2021	25.5	25.3	24.9
11/06/2021	23.6	23.1	22.0
17/03/2022	26.0	25.4	26.3
08/07/2022	21.8	21.7	20.1
04/11/2023	21.3	21.1	21.0

Table 7. Rat Island Marine Farm 1574, Site #2 water temperatures (°C). AAA Farm Site #2: Lat. 28°42.976'S, Long. 113°49.250'E.

Date	Surface	Mid	Bottom
18/05/2019	20.2	20.2	20.1
20/05/2020	22.1	21.6	21.5
30/07/2020	21.3	21.1	20.3
08/11/2020	21.5	21.6	21.7
22/03/2021	24.6	24.7	24.2
11/06/2021	22.1	22.4	21.9
16/03/2022	25.7	25.6	25.5
07/07/2022	21.6	21.5	21.1
03/11/2023	22.9	23.1	23.8

Table 8. Toxins and their biogenetic, microalgal origins*. Extracted from (FAO, 2011, pp. 6).

Toxin group	Abbreviation	Algae associated
Azaspic acid	AZA	<i>Azadinium spinosum</i>
brevetoxin-b	bTX	<i>Karenia brevis</i>
Domoic acid	DA	<i>Pseudo-nitzschia</i> spp.
Gymnodimine	GYM	<i>Karenia selliformis</i>
Okadaic acid	OA	<i>Dinophysis</i> spp.**; <i>Prorocentrum</i> spp.**
Palytoxin	PITX	<i>Ostreopsis</i> spp.**
Pectenotoxin-2	PTX	<i>Dinophysis</i> spp.**
Prorocentrolide	PCI	<i>Prorocentrum</i> spp.**
Saxitoxin	STX	<i>Alexandrium</i> spp.**; <i>Gymnodinium catenatum</i> , <i>Pyrodinium bahamense</i>
13-DM spirolide C	SPX	<i>Alexandrium ostenfeldii</i>
Yessotoxin	YTX	<i>Protoceratium reticulatum</i> , <i>Lingulodinium polyedra</i> , <i>Gonyaulax spinifera</i>

*For a complete list of harmful algae and associated toxins, see: www.marinespecies.org/hab/index.php

**Denotes the plural of species, i.e. several species of the indicated genus are reported to produce toxins from this group.

NB: Gymnodimine, pectenotoxin, prorocentrolide, spirolide, yessotoxin are considered of very little or no human health significance and are not regulated in Australia or New Zealand.

dominated phytoplankton samples through all seasons at the Abrolhos Island sites and exhibited significant blooms at the hottest period recorded at both sites (25–26 °C, March 2022). The shallower Pelsaert Island site displayed greater variability in the number of diatoms present and in accompanying water temperatures. Dinoflagellates (Dinophyceae) were present through all seasons at Rat Island but were absent or very scarce throughout a number of months at Pelsaert Island and especially when the *Oscillatoria* spp. (*Trichodesmium*) bloom occurred in May 2019.

Routine monitoring of phytoplankton (and other aspects) has been conducted by the Integrated Marine Observing System (IMOS) project at a number of reference stations around Australia and including off Ningaloo Reef since 2009 (Davies et al., 2022). Phytoplankton data includes abundance and biovolume measurements with various data sets such as raw and species (where these have been identified) available via the AODN portal.

Conclusion

Latitude Pearls and Abrolhos Aquaculture Australia have been monitoring environmental parameters at the Abrolhos Islands for many years and including before 2019 when an intensive program was conducted for a joint FRDC project. That information and data is publicly available through FRDC Report 2007/216 (Cropp et al., 2011).

In recent years, regular MEMP reporting for DPIRD has been conducted and as detailed herein, various heavy metal, marine microalgae/phytoplankton and biotoxins have been monitored since 2019. While the data has been targeted around the culture of the penguin oyster (*Pteria penguin*), much of it would also be useful for other species and for aquaculture management in the region generally, but particularly for shellfish.

The aquaculture companies herein were not seeking to harvest and sell whole shellfish, hence full WASQAP assessment was unnecessary (and adductor muscles are not included in WASQAP in any event) but merely to illustrate that heavy-metals and marine biotoxins are not a hazard for adductor muscle consumption. Some irregular presence (once above alert levels) of potentially toxic microalgae was detected in regional water samples but there was no evidence of any toxic effects on adductor muscles or whole shellfish from either the Rat Island marine farm site, or from Pelsaert Island, some 30 km away. In any event, only one exceedance of MBMMP limits was detected in 4 years (from 9 sample dates), with no toxins found. It therefore appears that there is minimal risk for the sale of penguin oyster adductor muscles, cultured at either site, for human consumption.

There appears to be a paucity of phytoplankton documentation for Western Australian waters, particularly the Abrolhos Islands area. This paper is

believed to be the first published study documenting the various phytoplankton occurring at the Abrolhos islands over time and their impact on cultured shellfish. The research has highlighted a further need to fully identify potentially toxic species to species level and then accurately determine whether they actually produce toxins at any time or are non-toxic, even though they may be very similar to and closely related to species of documented, toxin producing algae. However, complete species identification would require additional fresh samples, incur several more days analysis time and add significantly to the costs involved per sample. The financial viability as part of a regulated monitoring program would need to be considered by individual growers. There is a case in favour of obtaining such baseline information irrespective of the sampling program.

While the research work herein was not designed to be totally comprehensive in monitoring the seasonal presence of all species on a regular basis it does provide a snapshot of species diversity, abundance and variation over time. To fully understand the phytoplankton community and potential toxicity risks from species in the Abrolhos Islands area, a much more regular and structured sampling program, with associated toxin analysis, would be required, although the cost of such a program would be very significant. The EU Working Group (2019) has outlined the clear need to create effective sampling plans to allow the risks from PTP species to be properly assessed for any shellfish production area. Harvesting of whole shellfish from the Abrolhos Islands, such as scallops or oysters for human consumption, would require such a sampling program. The inherent variability in phytoplankton productivity, diversity and toxicity on a seasonal basis (as seen herein), are factors that must be incorporated in any plan if a comprehensive risk assessment is to be conducted. Compilation of monitoring results and input into the HAIS databases is expected to improve our knowledge and ability to manage the risks of harmful algal blooms into the future.

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Conflict of interest: The authors declare that they have no conflict of interest.

Author contributions: Derek Antony Cropp: Conceptualisation, methodology, visualisation, data analysis, writing, review and editing manuscript, project administration, supervision. Stuart Hellenen: Formal analysis, validation, data curation, review and editing, resources. Pia Boschetti: Marine farmer and funding acquisition, investigation. Murray Davidson: Marine farmer and funding acquisition, investigation.

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